HEALTH EFFECTS DIVISION
STANDARD EVALUATION PROCEDURE

DEVELOPMENTAL TOXICITY STUDIES

Health Effects Division
Office of Pesticide Programs
United States Environmental Protection Agency

October 11, 1993

STANDARD EVALUATION PROCEDURE

FOR DEVELOPMENTAL TOXICITY STUDIES

TABLE OF CONTENTS

ACKNOWLEDGEMENTS

Principle contributors to this document were David Anderson, Ph.D., Toxicology Branch 1; Stephen C. Dapson, Ph.D., Toxicology Branch 2; Roger Gardner, M.S., Toxicology Branch 1; and James N. Rowe, Ph.D., Toxicology Branch 2.

Quang Q. Bui, Ph.D., while working in Toxicology Branch 2, initiated compilation of the glossary and graciously provided the illustrations included in Section III. B. of this document.

Others who contributed significantly to the document were Gary Burin, Ph.D., formerly in the Science Analysis Branch; Deborah McCall, B.S., Toxicology Branch 2; and Karen Whitby, Ph.D., Toxicology Branch 2. Thanks are also extended to Hugh Pettigrew, Ph.D., Statistician in the Science Analysis Branch for his comments.

This Standard Evaluation Procedure is based in part on an earlier document published in June, 1985 entitled *Hazard Evaluation Division Standard Evaluation Procedure: Teratology Studies* prepared by Laurence D. Chitlik, Quang Q. Bui, Gary J. Burin, and Stephen C. Dapson (U.S. EPA, 1985).

Others who provided valuable comments on the document during its preparation included Karl Baetcke, Toxicology Branch 1, HED, U. S. EPA; Neil Chernoff, Developmental Toxicology Division, U. S. EPA; Mildred S. Christian, Argus Research Laboratories, Inc.; Thomas F. X. Collins, Mammalian Reproduction and Teratology Team, U. S. Food and Drug Administration; Marion Copley, Toxicology Branch 1, HED, U. S. EPA; Brian Demente, Toxicology Branch 1, HED, U. S. EPA; Saniju Diwan, Clement International Corporation; John Doherty, Toxicology Branch 1, HED, U. S. EPA; Penelope Fenner-Crisp, HED, U. S. EPA; Pamela A. Gilles, CIBA-GEIGY Corp.; Richard M. Hoar, Argus Research Laboratories, Inc.; Ronald D. Hood, The University of Alabama; Robert J. Kavlock, Developmental Toxicology Division, U. S. EPA; Carol A. Kimmel, Office of Research and Development, U. S. EPA; Pia Lindstrom, Clement International Corporation; Anthony K. Palmer, Huntingdon Research Centre, LtD.; Jennifer Seed, Toxic Effects Branch, Health and Environmental Review Division, OPTS, US S. EPA; Henry Spencer, Toxitology Branch 1, HED, U. S. EPA; Marcia Van Gemert, Toxicology Branch 2, HED, U. S. EPA; and Ashley Wickramaratne, Central Toxicology Laboratory, ICI Plc.

Contents

Contents

LIST OF FIGURES

LIST OF TABLES

Contents

I. INTRODUCTION

A. Using This Standard Evaluation Procedure (SEP)

The purpose of this SEP is to provide guidance on

- how to evaluate developmental toxicity data submitted to the Office of Pesticide Programs (OPP) to support registration or re-registration of pesticides and
- how to use that evaluation to support assessment of potential risks from human exposures.

This document addresses these purposes, and an appendix contains a glossary of terminology. Both parts of the SEP include reference material and tables of reproductive parameters that are essential knowledge in evaluation of developmental effects.

This SEP should be used in conjunction with the Pesticide Assessment Guidelines for developmental toxicity (Subdivision F, §83-3) and with guidance from senior scientists experienced in the review of developmental toxicity studies. *This SEP is not a rigid set of rules to follow in the evaluation of developmental toxicity studies.*

Criteria for data acceptability are described in Section II. Section III discusses biological and statistical principles used in evaluation of maternal and developmental toxicity end points as a basis for sound interpretations of developmental toxicity study results. Section IV discusses the use of developmental toxicity study reviews in the regulatory and risk assessment processes for pesticides that cause developmental toxicity in laboratory animals. Guidance is provided on writing the data evaluation record (DER) in Section V.

The glossary (Appendix) is a significant feature of this SEP. It includes definitions for terminology that is used to describe effects noted in developmental studies. The Appendix provides information on the developmental biology of species not commonly encountered in the review of developmental toxicity studies and supplements similar information on the developmental biology of the most frequently used species that is included in the main test of the SEP.

B. Working Definitions

The terms included in this section are frequently used in reports submitted to the Office of Pesticide Programs or can be found in the Agency's *Developmental Toxicity Risk Assessment Guidelines* (U.S. EPA, 1991b).

1. Developmental Toxicology

Developmental Toxicology is the study of adverse effects on the developing organism which may result from exposure prior to conception (either/both parents), during prenatal development (as in the FIFRA Guideline §83-3, Teratology Study) or postnatally to the time of sexual maturation (as in postnatal studies and FIFRA Guideline §83-4, Multigeneration Reproduction Study). Adverse developmental effects (i.e., developmental toxicity) may be detected at any point in the lifespan of the organism (see Table 1 for a summary of selected events for the most commonly used laboratory animals). Although the multigeneration reproduction study is more appropriate to the definition for developmental toxicology, the study protocols and end points of developmental toxicity considered in this SEP include: 1) death of the developing organism, 2) structural abnormality (malformations and variations), 3) altered growth, and 4) functional deficiency as they are evaluated by the protocol described in §83-3 of the FIFRA testing guidelines (see Section III. B., page 23).

2. Embryotoxicity and Fetotoxicity

Embryotoxicity and fetotoxicity are subsets of developmental toxicity referring to adverse effects on the developing conceptus prior to parturition. The distinguishing feature between the two terms is the stage of development during which the injury occurs. Effects occurring during the embryonic period are referred to as embryotoxicity, and effects occurring during the fetal period are referred to as fetotoxicity. Because the distinction between the two stages of development is arbitrary and therefore, subject to debate, and because both categories of effects are subsets of a broader type of toxicity, the term developmental toxicity is preferred instead of embryo- or fetotoxicity.

3. Teratogenicity

The 1982 Subdivision F Pesticide Assessment Guidelines (§83-3) define teratogenicity as follows:

> ...the property of a chemical that causes permanent structural or functional abnormalities during the period of embryonic development.

In the Guidelines for Developmental Toxicity Risk Assessment (U.S. EPA, 1991b), the term teratogenicity is defined as a "permanent structural change which may adversely

affect survival, development or function," (U.S. EPA, 1991b) which is consistent with the concept that teratogenicity is a type of developmental toxicity.

Table 1: Timing of selected events in the development of commonly used laboratory animal species and man (data compiled from NRC, 1989):

Organ System	Timing of event			
	Rat	Mouse	Rabbit	Human
Breeding age				
Male	8-12 wks.	8-10 wks.	6-7 mos.	14 yrs.
Female	8-12 wks.	6-8 wks.	5-6 mos.	13 yrs.
Duration of spermatogenesis[1]	48 d.	34-35 d.	48-51 d.	72-74 d.
Length of estrous cycle	4-5 d.	4-5 d.	polyestrus[2]	--
Length of menstrual cycle	--	--	--	28 d.
Duration of estrus	10-20 hrs	10-20 hrs.		--
Time of ovulation (after onset of estrus)	8-11 hrs.	2-3 hrs.	10 hrs.[3]	--
Gestation length	21-22	18-20	31-34	267-280
Stages of embryonic development				
2-cell	45 hrs.	24-38 hrs.	22-26 hrs.	
6- to 8-cell	79 hrs.	50-64 hrs.	32-40 hrs.	
Morula		68-80 hrs.	47-68 hrs.	
Blastocyst	107 hrs.	74-82 hrs.	68-76 hrs.	
Time of implantation	5.5-6 d.	4.5-5 d.	6-7 d.	6.5-7 d.
Period of major organogenesis	8-16 d.	6-15 d.	8-18 d.	19-80 d.
Weaning age	21 d.	21 d.	50 d.	

1 From p 55. in "Biologic Markers of Testicular Function." Ch. 4 in *Biologic Markers in Reproductive Toxicology*. National Research Council, National Academy Press, 1989.
2 Polyestrus is more than one estrus in a season.
3 Continuous estrous, with no regular cycle.

4. Structural Alterations

Structural alterations include both malformations and variations. A variation is a divergence within the usual range of structural constitution but which may not

adversely affect survival, development, or function. A malformation is a permanent structural change which may adversely affect survival, development, or function.

Because development is considered as a continuum of events, there is no universally accepted classification for many structural alterations as either variations or malformations (see illustrations beginning on page ? and Appendix on page 77 for examples of both malformations and variations). Other terminology that is used, but no better defined, includes anomalies and aberrations (which do not include variations) and deformations (which may be specific to alterations such as bone molding *in utero* that are mechanically induced).

5. Altered Growth

Altered growth is a change in offspring organ weight, body weight, size or delays in development such as delayed ossification. Changes in one end point may or may not be accompanied by signs of altered growth (eg., changes in body weight may or may not be accompanied by changes in such alterations as crown-rump length and/or skeletal ossification). Altered growth may be induced at any stage of development and may be permanent or reversible in nature. (Also see discussion beginning on page 27.)

6. Functional Developmental Toxicology

Functional developmental toxicology is the study of alterations or delays in functional competence of the organism or organ system following exposure to an agent during critical periods of pre- and/or postnatal development (see pages 5 and 45.

7. Risk Assessment

The NAS document on risk assessment (National Research Council, 1983) states that there may be four components in a risk assessment. These are (1) hazard identification, (2) dose-response assessment, (3) exposure assessment and (4) risk characterization. The Agency's *Developmental Toxicity Risk Assessment Guidelines* (U.S. EPA, 1991b) combine the first two concepts into a *hazard identification/dose response evaluation*, and describe it as follows:

> Hazard identification/dose-response involves the evaluation of all available experimental animal and human data and the associated doses, routes and durations of exposure to determine if an agent causes developmental toxicity in that species and under what exposure conditions.

The Agency's Guidelines state that the *exposure assessment* defines populations exposed to the agent in terms of their magnitude, duration, schedule and route of exposure. Because fetal exposure depends on conditions of maternal exposure and pharmacokinetics of the pesticide (i.e., whether the pesticide is absorbed and reaches the fetus), it is assumed that, in most cases, a single exposure generally at a maternally toxic dose at the critical time in development is potentially sufficient to produce an adverse fetal effect.

The final component, *risk characterization*, is described in the Risk Assessment Guidelines as follows:

> ...(R)isk characterization involves integration of the toxicity information from the hazard identification/dose-response evaluation with the human exposure estimates and provides an evaluation of the overall quality of the assessment, describes risk in terms of the nature and extent of harm, and communicates the results of the risk assessment to a risk manager.

(Also see Section IV, page 61.)

8. Study Designs

a. The Principle Study

In the developmental toxicity study described by §83-3 of the Subdivision F Guidelines, pregnant animals are randomly assigned to each of at least four test groups. Each of three treatment groups then receives a different level of the test substance during the period of major organogenesis. Events potentially affected by administration of a test substance during the period of major organogenesis are summarized in Table 12 on page 67. Near the end of gestation the animals are sacrificed, and each treated group is compared to a concurrent control group with regard to the end points described in Section III. below. This protocol is similar to the Food and Drug Administration's (FDA) Segment II study protocol.

b. Postnatal Studies

Postnatal developmental toxicity studies evaluate the potential effect of pesticides on the structure and function of organ systems in offspring after birth. Pesticides causing neurotoxicity in adults and those that cause central nervous system (CNS) malformations are required to be evaluated for their potential effect upon the structure and functioning of the nervous system in offspring exposed during pregnancy and lactation (*Pesticide Assessment Guidelines, Subdivision F, Hazard Evaluation: Human and Domestic Animals*, Series 81, 82, and 83, *Neurotoxicity*, 1991a).

Postnatal evaluation of development in other organ systems in addition to the nervous system may be submitted for review, and those studies will report on end points other than those listed in Table 5 (page 46). For example, when the compoundhas a hormonal action, a special protocol is designed to evaluate postnatal effects on the development of endocrine structures and functions (e.g., anogenital distance, nipple development in male pups, residual Wolfian ducts, etc.). Since the Agency's guidelines are recent, and only a few postnatal neurotoxicity studies on pesticides have been submitted and are under review, this SEP can not provide much discussion of these studies beyond that found in the Agency's risk assessment guidelines (U. S. EPA, 1991b).

With regard to the developmental neurotoxicity study, the test substance is administered in the rat from gestation day 6 through day 10 of lactation. Dosing (usually oral) is not performed on the day of parturition in animals that have not completely delivered their offspring. The neurotoxicity evaluation includes observations to detect neurologic and behavioral abnormalities, determination of motor activity, response to auditory startle, assessment of learning, neuropathological evaluation, and brain weights. This type of study may be either a separate, follow-up to a standard study and/or adult neurotoxicity study, or as a part of a multi-generation reproduction study.

c. Range-finding Studies

The range-finding study is designed to establish a basis for selection of dose levels to be used in the principal (§83-3) developmental toxicity study. If minimal effects are observed in the principal study, a range-finding study should be available to verify that the doses tested at least approached a toxic level. These preliminary studies generally lack the critical internal fetal examinations (i.e., skeletal and visceral) found in a principal study and have insufficient numbers of litters to fully assess potential maternal or developmental toxicity. Therefore, range-finding studies should not be used to establish no-observed-effect levels (NOEL).

In the absence of range-finding studies, acute oral toxicity and metabolism studies in the same strain of animal may also provide useful information for setting dose levels. However, single dosing and differences between pregnant and non-pregnant animals in pharmacokinetics and metabolism can cause errors in the selection of an appropriate dose range and in the interpretation of the principal effects produced.

C. When Developmental Toxicity Studies Are Required

In general, the Office of Pesticide Programs requires developmental toxicity testing in two species to support the registration of a product intended for food use (i.e.

when tolerances or exemptions from tolerances are considered) and for nonfood uses if significant exposure of human females of childbearing age may reasonably be expected. Temporary tolerances generally require testing in only one species. In thecse of a pesticide that may be structurally similar to known developmental toxicants, there may be a need for developmental toxicity studies when not otherwise required.

When developmental toxicity studies fail to show maternal or fetal effects, additional testing may not be necessary if the "limit test" requirements (a dose level of 1000 mg pesticide per kg body weight or more) in Subdivision F are met or exceeded. If the doses tested are lower than the limit dose and no maternal or developmental effects are observed, an additional study at higher doses can be recommended when a study in a second species does not show maternal or fetal effects.

The Agency's Risk Assessment Guidelines (U.S. EPA, 1991b) note that the minimum data base for identification of a potential developmental hazard associated with a chemical is described on page 61.

II. DATA ACCEPTABILITY

The potential of a pesticide to cause developmental toxicity is assessed from studies conducted under good laboratory practices (GLP; 40 CFR §160.185) and sound scientific principles. In order to reduce errors in the risk assessment process, data from studies that do not conform to GLP should be rejected when protocol deviations compound the results or their interpretation beyond the intent of Subdivision F, §83-3 testing guidelines.

The Health Effects Division has conducted a rejection rate analysis (HED, 1993) to determine the factors that have most frequently caused toxicological studies required for reregistration to be rejected (thus delaying the review process). The rejection analysis was confined to studies that were considered upgradable or unacceptable for regulatory purposes (including supplementary studies with "fatal" deficiencies that were not upgradable and invalid studies). The classification system used in the Toxicology Branches to "grade" studies is described in Section II. C. on page 17.

Rejection factors indicated that additional information was needed on a study protocol to upgrade the study. These items included descriptions of the following:

(1) the test substance (purity, composition, stability, etc),
(2) preparation techniques for the test substance,
(3) dosing dates,

(4) source of the test animals,
(5) randomization procedures,
(6) mating methods (artificial insemination, mating records, etc),
(7) sacrifice order,
(8) examination procedures (fixing and staining techniques for examination of uteri, fetuses, etc.), and
(9) statistical methods and results of analyses.

These principles are the basis for this section's discussion of the characteristics of maternal and developmental toxicity end points and their relationships to each other in the interpretation of study results.

Studies have also been rejected because of reporting deficiencies such as thefollowing:

(1) appendices containing individual animal (maternal or fetal) data,
(2) summary tables,
(3) gravid uterine weights,
(4) fetal incidences without litter data, or litter data without fetal results, and
(5) the litter was not the experimental unit and further statistical analyses were needed.

The rejection analysis also indicated that the review of studies has been delayed because historical control and range-finding data were not available to aid in the interpretation of a developmental toxicity study's results. Finally, factors that caused a study to be classified as unacceptable and required asking for another test included the following:

(1) doses selected in the studies did not produce toxicity such that maternal and/or developmental NOELs could be established;

(2) excessive maternal toxicity precluded the availability of an adequate number of litters for examination; and
(3) unconventional assessments for skeletal or visceral examinations were used.

A. Acceptability for Review

Information to be included in the report of a developmental toxicity study is described in *Subdivision F: Hazard Evaluation: Humans and Domestic Animals*, §80-4 and §83-3 and the FIFRA Accelerated Reregistration Phase 3 Technical Guidance document (U.S. EPA, 1982; U.S. EPA, 1989). The guidelines are the basis for 16 Acceptance Criteria used in the Health Effects Division for determining the acceptability of a developmental toxicity study for review. The criteria should not be used as the basis for a final conclusion regarding scientific or regulatory acceptability

unless they indicate the study is invalid or can not be upgraded. The Acceptance Criteria are listed as follows:

(1) Technical form of the active ingredient tested (see Section II.A.1.a. and b.).
(2) At least 20 litters/dose group for mice, rats, or hamsters are available. At least 12 litters/dose group for rabbits are available (three test groups and control group) (see Sections II.A.2.a. and II.B.2.a. and b.).
(3) At the high dose, maternal effects are reported as significant (or a limit dose is tested, 1000 mg/kg/day) (see Section II.A.2.a. and III.A.). A study may also be accepted if the highest dose tested is associated with developmental toxicity in the absence of maternal toxicity.
(4) At the low dose, no developmental toxicity is reported (see Section II.A.2.b.).
(5) Dosing duration is at least during the period of major organogenesis but may extend up to one day prior to term.
(6) Analysis for test material stability, homogeneity and concentration in dosing medium.
(7) Individual daily observations.
(8) Individual body weights.
(9) Individual food consumption (not required in §83-3 guidelines, only necessary when the test substance is administered in the diet).
(10) Necropsy on all animals.
(11) Individual uterine examination including number of fetal deaths, early and late resorptions and numbers of viable fetuses per sex (see Section III.B.6.).
(12) All ovaries examined to determine number of corpora lutea (except when the test species is the mouse).
(13) Individual litter weights and/or individual fetal weights/sex/litter.
(14) Individual fetal external examination.
(15) Individual fetal skeletal examination for 1/3 to 1/2 of each litter for rodents and all fetuses for rabbits.
(16) Individual fetal soft tissue examination.

Exceptions to these criteria can be made on a case-by-case basis, but they are usually for special studies. Once a study has met these criteria, it should be evaluated according to the principles discussed in Section III. below.

1. Test Substance

a. Technical grade active ingredient and impurities

As stated above, the technical form of the active ingredient is usually administered to the pregnant animal. As required by the EPA GLP's (Section 40 CFR, 160.185), the test compound description should include the name, chemical abstracts (CAS) number, strength, purity, composition and stability under the conditions of administration. Where there are "significant" levels of impurities (either defined by >0.1% or by toxicity, if known), the reviewer will have to evaluate the potential contribution of these impurities to any developmental effects which might be observed. For example, dioxins, impurities found in chlorophenoxy herbicides (e.g.,

2,4 D or 2,4,5-T), and nitrosamines, found in cutting fluids, (Murphy, 1980; Harbison, 1980, p. 164) are known developmentally toxic agents which have presented considerable concern to the Agency in the past.

In some cases, the formulated product may be used. For example, when the physical form of the active ingredient is not soluble in commonly used vehicles (see Section II. B. 2., page 14) or when the purpose of the study of a test substance with a known developmental hazard is needed to assess potential risks associated with dermal or inhalation exposures, studies with formulations will be submitted for review.

b. Physical form of the test substance

Pesticides may range in physical characteristics from gases/vapors, to liquids, amorphous powders or crystals. The physical state of the test material is an important factor in determining the method of administration for hazard identification and the most likely route of exposure in risk assessment (see page 14). For example, solid test material must be finely ground if insoluble in the vehicle (see discussion on page 16).

When dermal developmental toxicity studies are reviewed, skin irritation must be considered since it may affect the integrity of the dermal layer and thus potentially enhance dermal absorption of the applied pesticide (Kimmel and Francis, 1990). The *Guidelines for Developmental Toxicity Risk Assessment* also note:

> ...absorption data are needed, both when a dermal developmental toxicity study shows no developmental effects, as well as when developmental effects are seen. The results of a dermal developmental toxicity study showing no adverse effects and without blood level data (as evidence of dermal absorption) are potentially misleading and would be insufficient for risk assessment, especially if interpreted as a "negative" study. (see Section IV.A.2.b., page 65)

2. Dose levels

a. Highest dose

Dosing at levels that produce excessive toxicity (i.e., death, abortions or total litter resorptions) may result in an unacceptable number of litters for evaluation in treated groups (<20 for mice, rats and hamsters; <12 for rabbits). The study with an excessively toxic highest dose level could be rejected if results at lower dose levels do not define an NOEL or LOEL.

On the other hand, an oral gavage study might be rejected if the highest dose has no maternal or developmental effects and is less than 1000 mg pesticide per kg

body weight per day (limit dose). Selection of limit doses for studies by other routes (e.g., inhalation, dermal etc.) should be considered on a case-by-case basis. For example, a dermal study without maternal or developmental toxicity and withoutevidence of absorption of the test substance through the skin are not sufficient for regulatory purposes (see page 16).

b. Lowest dose

The Subdivision F Guidelines recommend, "...the lowest dose level should not produce any evidence of toxicity." If maternal toxicity is the only finding at this dose level, the study may still be considered acceptable when a study in the second species has a lower dose level without maternal toxicity (see page 62).

There may be evidence that the test animal is uniquely sensitive during pregnancy to a test substance, and therefore, a maternal NOEL would be required. With suitable interpretation, multigeneration reproduction studies may aid in determining the maternal no observed effect level in rats since both developmental and reproduction studies are generally available for a given test substance in that species. However, differences in route and duration of dosing may influence the expression of the test substance's maternal toxicity in the multi-generation and developmental toxicity studies, and the reviewer should consider possible differences in absorption, distribution and excretion associated with those factors when using the two studies to determine a maternal NOEL.

3. Individual Animal Data

The report must include individual animal data so that the fetal effects observed may be related, whenever possible, to the condition of the animal bearing affected fetuses. Soft tissue, skeletal, and external alterations may comprise a syndrome or consistent pattern in individual fetuses which may be seen in reports on individual animals. In addition, the report must include summary data (number and percentage) for each group on both litter and fetal data. (Also see Section III. C., page 48.)

B. Evaluation of Study Conduct

After a study has been accepted for review, its conduct is evaluated.

1. Test Species

Generally, rats and rabbits are the preferred test species for standard testing of pesticides. Studies conducted in mice or hamsters may also be acceptable. However, alternatives to rat and rabbit must be justified.

Evaluation of Study Conduct

a. Reproductive Status of Animals

Virgin females should be selected for testing since confirmation of pregnancy in parous dams cannot be accurately determined. Furthermore, if corpora lutea are counted for the determination of pre-implantation loss, nulliparous animals are preferred (not in bibliography, WHO, Technical Report Series No. 364, 1967 ; See discussion on page 13).

i. Pregnancy Rate

The pregnancy rate (# pregnant/# mated) is an important index for assessing the adequacy of reproductive performance of the test animals selected. A low pregnancy rate will limit the number of litters available for examination and may require initial group sizes larger than those recommended in the §83-3 guidelines so that the recommended number of litters will be available at the end of the study.

Low pregnancy rates may suggest maternal health problems, poor animal husbandry, or may be due to dosing prior to completion of implantation. Other common causes for reduced pregnancy rate include immaturity, absence of breeding experience and genetically-generated testicular atrophy and aspermia. Regardless of the mechanism by which the pregnancy rate is decreased, a reduction in available litters will result in a less sensitive evaluation of potential developmental toxicity.

Both mating and artificial insemination have been used in rabbit developmental toxicity studies. Artificial insemination presumably will ensure a knowledge of the exact time of insemination (Gibson et al., 1966). However, a high percentage of pre-implantation loss and small litter size have been described with this method (Woo and Hoar, 1982; Woo, 1984). A pregnancy rate of 80% and greater in rabbits is usually obtained by artificial insemination (Gibson et al., 1966, Adams, 1961; Woo, 1984) If the number and concentration of sperm used in the inseminating procedures are adequate (1 million sperm; Walton, 1927; Gibson et al., 1966). Artificial insemination techniques, semen dilution factors, and sperm motility are a few of the important parameters that should be included in the report. The time between administration of luteinizing hormone, generally HCG (human chorionic gonadotropin), and insemination is also critical since ova lose viability within 12 hours after hormonal treatment (Chang, 1958). High pre-implantation loss also reflects variations in administered HCG dose and interaction of the ovulatory reflex and the hormonal response, against the baseline response.

Dosing in developmental toxicity studies is initiated close to the time of implantation as recommended in the FIFRA Guidelines (§83-3), and no significant differences in pregnancy rates between any test groups and controls should be

observed. The time of implantation may vary (see Table 1 on page 3), and dosing does not always begin after implantation. If significant treatment-related differences are observed, the study should be evaluated to verify that sufficient numbers of litters are available for an examination of all required parameters. The study on reproduction (Guideline §83-4) is more appropriate in elucidating compound-related alterations in pregnancy rate.

ii. Corpora Lutea

The number of corpora lutea from both ovaries should be counted. Corpora lutea are evidence of released ova and usually outnumber implantation sites. Virgin females are preferred in developmental toxicity studies to prevent potential counting error when distinguishing between corpora lutea and corpora albicantia (from earlier ovulations).

Theoretically, the number of corpora lutea among test groups should be similar, since administration of test compound is to be performed after implantation has been completed.

iii. Pre-implantation Loss

Pre-implantation loss is determined as the difference between the number of corpora lutea and implantation sites. An increase in pre-implantation loss suggests a time-dosing error (dosing prior to the completion of implantation), which results in compound induced and/or maternal stress-related embryo lethality (see pages 12 and 27). However, very early implantation loss, day 6 or 7, may not leave an implantation scar (metrial gland).

High pre-implantation loss may limit the sensitivity of a study because the number of implants left to evaluate end points of developmental toxicity is decreased. Implantation times for selected species are listed in Table 1 on page 3.

b. Diseases of Laboratory Animals

Pasteurellosis and coccidiosis are common diseases encountered in rabbits which may be manifested as: nasal discharge, diarrhea, and congested lungs, as well as pitted kidneys and brain lesions observed at necropsy (Benirschke et al., 1978). These observations are similar to those that may result from toxicity. A generally high or non-dose-related incidence of such observations is suggestive of a questionable health status of the test animals. A study should be rejected if the disease interferes with determining NOELs for maternal and developmental toxicity.

In some studies, especially with rabbits, the incidences of diseases may be reduced by treatment with antimicrobial agents prior to studyinitiation. Use of disease-free animals or quarantining (as recommended in GLP and §83-3 guidelines) prior to initiating a study may reduce possible effects of treatment with antimicrobials on development. Studies in which measures are used to reduce the incidence of infections should be considered acceptable on a case-by-case basis, depending upon circumstances of the quarantine period, type of antimicrobial agents administered to test animals and other factors as they affect developmental and maternal toxicity end points.

2. Route of Administration and Dosing Medium/vehicle

Absorption is an important consideration in evaluating appropriateness of the vehicle and route of administration of a test substance. The Agency's risk assessment guidelines (U.S. EPA, 1991b) provide the following guidance on absorption:

> ...absorption data in laboratory animals for studies conducted by any relevant route of exposure may assist in the interpretation of the developmental toxicity studies in the animal models for the purposes of risk assessment...absorption data are needed both when a dermal developmental toxicity study shows no developmental effects, as well as when developmental effects are seen. The results of a dermal developmental toxicity study showing no adverse developmental effects and without blood level data (as evidence of dermal absorption) are potentially misleading and would be insufficient for risk assessment, especially if interpreted as a "negative" study. In studies where developmental toxicity is detected, regardless of the route of exposure, absorption data can be useful to establish the internal dose in maternal animals for risk extrapolation purposes.

a. Oral studies

Dosing by gavage is the preferred route of administration in developmental toxicity studies because this route is similar to the most frequently encountered route of human exposure (dietary). Studies conducted by the dermal or inhalation routes may also be appropriate since these routes may be comparable to the primary routes of human exposure for the conditions of a pesticide's use. Dietary administration may also be appropriate if maternal toxicity is adequately demonstrated or if that route changes the manner in which the animal metabolizes the test substance.

Optimally, the vehicle selected for the administration of the pesticide should not interfere with absorption of the test material or induce maternal or developmental toxicity in test animals. Actually, all vehicles affect, in some manner, the absorption of a material either in the gastro-intestinal tract or on the skin. If the toxicity of the vehicle is unknown, results from a vehicle control group should be and usually are available for consideration. Commonly used vehicles are listed in Table 2.

When a substance is insoluble in water, corn oil or carboxymethycellulose may be used to form suspensions and/or solutions. For example, when corn oil is used, the volume administered should be limited to 3-4 ml/kg; a larger amount can cause

Table 2: Examples of vehicles frequently administered by gavage (oral) or dermally in developmental toxicity studies *

Name	Route	Comments
Acetone	dermal	Neurobehavioral toxicity; repeated dermal use leads to defatting of skin; oral not preferred.
Carboxymethyl-cellulose (CMC)	oral	Practically inert polymer cellulose (CMC).
Corn oil	oral, dermal	Energy source (can alter food consumption, b.wt.); can act as a laxative.
Dimethyl sul-foxide (DMSO)	-----	Repeated dermal use can defat skin; repeated oral exposure can produce corneal opacities; may enhance absorption.
Ethanol	dermal	Oral exposure can produce developmental effects.
Glycerol; glycerin	oral, dermal	Energy source; absorbs moisture from air.
Gum arabic; acacia	oral	Virtually inert biologically.
Methyl cellu-lose/methocel	oral (mixture)	Can act as laxative.
Polyethylene Glycol-400/ Carbowax	oral	Does not hydrolyze or deteriorate on storage; will not support mold growth (water soluble emulsifying/dispersing agent).
Saline	oral, dermal	No limitations unless test substance is not water soluble.
Water	all	Vehicle and solvent of choice unless test substance is not water soluble.

* Adapted from S.C. Gad and C.P. Chengelis (1988). Chapter 10-Routes, Formulations and Vehicles, Appendix C, Acute Toxicology Testing Perspectives and Horizons, The Telford Press; vaginal, rectal, subcutaneous, intramuscular and intravenous uses have been deleted from the uses since these would not be generally appropriate for developmental toxicity studies

severe diarrhea which causes the test substance to be excreted from the animal before its absorption can be accomplished. Corn oil or mineral oil may also sequester lipophilic agents which may give spurious negative results (Gad and Chengelis, 1988). In studies showing no toxic effects, the possibility of the use of an inappropriate

vehicle should be considered, particularly when clinical signs such as diarrhea are observed with equal frequency in vehicle control and treatment groups show no dose-response relationship and doses of the test substance exceed expected toxic levels indicated by other toxicity studies.

Gels and resins present problems because of their viscosity characteristics at room temperature. Warming may alleviate this problem, but thermal degradation of the test substance may occur. Dissolution characteristics of solid dosage forms, which depend on formulation in addition to the properties of the chemical itself must be considered (e.g., vehicles may decrease the permeability of a suspension or capsule to water and retard dissolution and diffusion) (Gad and Chengelis, 1988).

b. Dermal studies

The dermal route of administration in a developmental toxicity study without toxicity of any kind or without data demonstrating bioavailability to the fetus may beof limited value for assessing the potential of a pesticide to pose a developmental hazard. Dermal developmental studies in which marked skin irritation is observed are also unacceptable since the skin effects may complicate the conduct of the study or interpretation of its results and since human exposure situations might increase protection against the irritation hazard. For those pesticides that are capable of readily penetrating the skin of laboratory animals, the conventional dosing period (during

There are solvents such as acetone that may be appropriate for hazard identification because they promote abosroption through the skin (Kimmel and Francis, 1990), but these substances may not be appropriate vehicles for a study with risk characterization as its purpose because they will not be a part of the material to which human skin will be exposed. The ideal solvent for the dermal study should mimic human exposure and include no or low systemic toxicity, rapid evaporation, no skin irritation, chemical nonreactivity and solubility that maximizes absorption of the test chemical (Kimmel and Francis, 1990). However, certain vehicles (solvents) are inappropriate, e.g., DMSO, ethanol or glycol ethers because there may be toxicity toxicity associated with their use.

c. Inhalation studies

For inhalation studies, the critical aspects of dosing include respirable particle sizes if the test substance is a solid or liquid material and limit concentration for any test substance regardless of physical state. The current criteria for particle size (U. S. EPA, 1988) is:

> ...at least 25% of the particle distribution used in these studies should be in the submicron range for acute and repeat exposure studies.

and limit concentration is defined in §81-2 and §82-4 of the FIFRA test guidelines as:

> ...5 mg/l (actual concentration of respirable substances) for 4 hours or, where this is not possible due to physical or chemical properties of the test substance, the maximum attainable concentration...

It should be noted that these criteria are being reconsidered and an appropriate HED scientist should be consulted about acceptable exposure conditions for dosing in inhalation developmental toxicity studies.
major organogenesis) may be appropriate, but for those with poor dermal penetration, a prolonged dosing period from just after mating may be more appropriate.

C. The Core Classification System

The core classification system is a procedure by which studies are "graded" with regard to the adequacy of methods, reporting, and other factors discussed in Section II. page 8, and it does not consider the scientific results beyond the intent of §83-3 testing guidelines (Engler and Quest, 1988). The four categories are defined by Engler and Quest as follows:

> **Core Guideline:** the study totally conforms with Subpart F Guidelines (§83-3).
>
> **Core minimum:** the study is sufficient to fulfill the intent of the Guidelines.
>
> **Core supplementary:** the study does not meet minimum criteria even though it may contain scientifically useful information.
>
> **Invalid:** the study is essentially useless with respect to scientific information as well as for regulatory purposes.

Studies classified as guideline or minimum as well as supplementary studies that have been upgraded are considered to be acceptable for regulatory purposes whereas those given classifications of supplementary (and not upgradable) or invalid are unacceptable in most cases.

III. INTERPRETATION OF RESULTS

Interpretation of results from developmental toxicity studies depends on an understanding of the biological and statistical nature of the end points observed. Wilson (1965, 1973, and 1977) developed generalizations from the body of accumulated experience with animal and human observation that should be the basis

of any interpretation of developmental toxicity data. These principles are summarized as follows:

> Susceptibility to developmental toxicity depends on the genotype of the conceptus and the manner in which this interacts with adverse environmental factors (see page 23).
>
> Susceptibility to developmental toxicity varies with the developmental stage at the time of exposure to an adverse influence (see Section III. B., page 23).
>
> Toxic agents act in specific ways (mechanisms) on developing cells and tissues to initiate sequences of altered developmental events.
>
> If a critical dosage reaches or is able to accumulate in the developing tissues, developmental alterations will occur.
>
> The four manifestations of developmental toxicity are death, structural alterations, growth retardation, and functional deficits (see discussion beginning on page 23).
>
> The manifestations of developmental toxicity increase in frequency and degree with increasing dosage from the no-observed-effect (NOEL) to the totally lethal level.

A. Maternal Toxicity End Points

The determination of maternal toxicity in teratology studies should be compared to observations from other toxicological tests to establish differences in response during pregnancy and adequacy of the doses tested (see Section II. B. 1. b. i.). Maternal toxicity end points commonly encountered in developmental toxicity studies are listed in the Agency's *Developmental Toxicity Risk Assessment Guidelines* (U. S. EPA, 1991b) and summarized in Table 3 (page 20).

1. Mortality and Clinical Observations

Range-finding and other toxicity studies conducted prior to the principal developmental toxicity study should provide information on the clinical signs of toxicity characteristic of the test substance. On a case-by-case basis these range-finding studies may be used to support equivocal evidence of maternal toxicity in a main study (see page 60). In these cases the range-finding studies must be included in the Data Evaluation Record (DER) or another DER prepared on the range-finding study. Other factors such as disease, environmental conditions (e.g., variations in

housing conditions, temperature, humidity, light-cycle caging, etc.) and technical errors may also produce mortality or clinical signs similar to those observed in animals treated with the test substance and these factors must be considered along with the background information from the preliminary studies in an evaluation of the results.

The cause of death should be identified from the necropsy data if possible. Incidences of congested lungs, reddening of the tracheal lining, fluid accumulation in the lungs are suggestive of either gavaging error or disease. Consequently, maternal death due to causes other than pesticide toxicity should always be considered with regard to this affect on the adequacy of the study.

Decreases in mating or fertility may be important indicators of maternal health and the general status and suitability of the strain for use in toxicity testing. When dosing is initiated prior to cohabitation, mating and fertility are important indicators of toxicity. Changes in the gestation length where studied appropriately may indicate effects on parturition and/or hormone homeostasis, while decreases in postnatal viability may possibly reflect maternal and/or developmental toxicity.

A dose-response relationship should always be evident before clinical signs and mortality are attributed to administration of the test compound. Where a dose-relationship is not clear, the effects of disease, environmental conditions, or technical errors should be eliminated as possible causes of the observed mortality and clinical signs. Clinical signs, organ weights, and/or pathology become very important when testing pesticides which result in very little body weight gain effects prior to death. However, the developmental toxicity study protocols considered in this SEP often do not include observations such as organ weights and histopathology, and the highest dose group may be the only group with increased clinical signs or mortality. In this case, a statistically significant pairwise difference between the control and high dose groups and a statistically significant trend as well as comparisons of the signs reported in the developmental toxicity study with signs observed in other studies in the same species should be used to establish the observations as compound-related.

2. Maternal Body Weight

Animals are often randomly assigned to groups based on evenly distributing females by body weight. If body weight is the criterion, this assignment evenly distributes the variation in the initial body weight among the groups. When this assignment is by a specified randomization procedure (e.g., blocked randomization), initial mean body weights and their variances become more homogeneous. The body weight gain of all the groups prior to dosing should be comparable. If they are not, the predosing differences should be accounted for in an analysis of subsequently observed weight gain differences (see pages 56 and 58).

Maternal Toxicity End Points

Table 3: End Points of Maternal Toxicity Usually Encountered in Developmental Toxicity Studies

End point	Comments
Mortality	When not due to gavaging error or other problems incidental to the study.
Body weight	Day of study initiation, from initial day of dosing up to and including day of sacrifice.
Body weight change	From day of study initiation to day dosing begins, during dosing (increments of time during dosing), from first day post-dosing to day of sacrifice.
Food & water consumption	When available.
Clinical evaluations	Types, incidence, degree and duration of clinical signs. Examples are listed below: a. Anogenital staining b. Definitive decreases in defecation c. Uterine bleeding d. Ataxia or tremors e. Biologically significant signs of somnolence or anesthesia f. Prostration g. Abortions (may also indicate developmental/ reproductive effects. (Objective measures should be emphasized.) Enzyme markers (reserved pending evaluation of cholinesterase inhibition assessment workgroup). Clinical chemistry results.
Gestation length	Useful when pregnant animals are allowed to deliver, but requires specific monitoring.
Gross necropsy and histopathology	When available (Should be encouraged when body weights are unaffected.).

Body weight and changes in body weights should be carefully considered as indicators of maternal toxicity. The risk assessment guidelines (USEPA, 1991b) note:

> Body weight and the changes in body weight are viewed collectively as indicators of maternal toxicity for most species,... Body weight changes may provide more information than a daily body weight measured during treatment or during gestation. Changes in weight

gain during treatment could occur that would not be reflected in the total weight change throughout gestation, because of compensatory weight gain that may occur following treatment but before sacrifice. For this reason, changes in weight gain during treatment can be examined as another indicator of maternal toxicity.

Changes in maternal body weight corrected for gravid uterine weight at sacrifice may indicate whether the effect is primarily maternal or intrauterine. For example, a significant reduction in weight gain throughout gestation and in gravid uterine weight without any change in corrected maternal weight gain generally would indicate an intrauterine effect. Conversely, a change in corrected weight gain and no change in gravid uterine weight generally would suggest maternal toxicity and little or no intrauterine effect. An alternate estimate of weight change during gestation can be obtained by subtracting the sum of the weights of the fetuses. However, this weight does not include the uterine or placental tissue, or the amnionic fluid.

Determination of maternal toxicity in the rabbit is more difficult than in the rodent. Rabbits spontaneously stop eating periodically during gestation which results in random body weight reductions. These random body weight reductions decrease the reliability of body weight as an indicator of maternal toxicity (Kimmel and Price, 1990). In cases of minimal body weight decrements, range-finding studies at higher dose levels may provide additional information on a test substance's effects on maternal body weight.

Statistical analysis selected by investigators for body weight gains in rabbits varies from study to study. Inspection of individual animal data may indicate a dose or a compound related increase in the number of animals with body weight gain decrements of one standard deviation or more below group means for the groups under consideration. This may indicate that the reduced body weight gain decrement across groups was not a random event, but was statistically significantly greater in the highest dosed group. A suitable statistical test, such as Bartlett's test for homogeneity of variances (see page 56), could be used to verify that distribution of animals with low weight gains among the test groups is not random. If the preliminary statistical evaluation confirms nonrandom distribution of data or heterogeneity of variances, the data can be transformed to stabilize variability and be reevaluated, outliers (if only one or two are present in each group) can be removed, or non-parametric tests such as the Kruskal-Wallis test or the Wilcoxon-Mann-Whitney U test can be used (see Section III. E., page 55 and Table 10, page 58). However, statistically significant differences in group mean body weight gains should be considered with respect to their biological significance. Rabbits in teratology studies weigh between 2.5 and 3.5 kg, and body weight changes are often one or two per cent of the maternal body weight, which may be indicative of nothing more than reduced food consumption.

3. Food and Water Consumption

Reporting food and water consumption data is not presently required by the Subdivision F Guidelines for developmental toxicity studies (§83-3). In those studies using the dietary or drinking water routes for administration of the test material, food and water consumption are used to calculate the dose. Food and water consumption data may be useful for assessment of maternal effects regardless of the route of administration since decreases in consumption may lead to decreases in body weight or indicate decreased palatability of the test diet in dietary studies. Small changes in water consumption, even if statistically significant, are difficult to evaluate, but if such decreases are dose- or compound-related they may indicate effects on excretory function, appetite, or water consumption.

Determining the efficiency of food utilization when food consumption is affected by the test material may be helpful in characterizing maternal toxicity, but these results are subject to the variability associated with the food consumption and body weight components of the calculation. Efficiency of food utilization (E_f) can be calculated for time t as follows:

$$E_f = \frac{grams\ body\ weight\ change\ per\ unit\ time}{grams\ food\ consumption\ per\ unit\ time} \times 100$$

This calculation gives the percentage efficiency with which the animal converts food for maintenance. Low efficiency compared with controls indicates toxicity in the consuming animals.

4. Organ Weights and Enzyme Markers

At the present time (with the exception of the gravid uterine weight), neither organ weights nor enzyme markers are required by Subdivision F Guidelines and as a result the data are seldom available for evaluation. These data may be submitted in support of maternal toxicity, especially in cases where the test substance causes little or equivocal body weight decreases but is associated with target organ toxicity. Organ weight changes supported by histopathology may indicate maternal toxicity in the rodent or the rabbit. For example, liver pathology in a chronic study which is associated with absolute and relative liver weight increases may support maternal toxicity if these changes are reported in the developmental toxicity study. Enzyme markers may be used when supported by histopathology from other studies. The use of cholinesterase inhibition in plasma, erythrocytes, and the brain in developmental toxicity studies has been used to characterize maternal toxicity, but further comment is reserved pending results of a work group on cholinesterase inhibition.

B. Developmental Toxicity End Points

Because the maternal animal, and not the conceptus, is the individual treated during gestation, data generally are calculated as incidence per litter or as number and percentage of litters with particular end points (USEPA, 1991b). Developmental toxicity end points considered by the Agency are listed in Table 4 on page 24.

Variability in the response of each individual, strain, or species to the test substance is an important influence on fetal observations in developmental toxicity studies. Individual maternal differences in absorption, distribution, metabolism, and excretion of a test substance can vary the level of exposure of individual fetuses from different litters within a test group. Another source of variability in the response of individuals, strains, or species is the numbers of corpora lutea and implantations present. These two parameters initially determine the total number of fetuses at risk in each litter and are examples of the genetically determined factors that affect susceptibility of fetuses to developmental toxicity.

The stage of development affected by a test substance is an important factor in the nature of the developmental toxicity induced. Since true differentiation in the earliest stages of embryonic development (other than cell positions relative to surfaces and other cells in the early embryo, see Table 1) has not occurred, and since embryonic cells probably have not developed specific roles, sensitivity of one or a few cells to a test substance's toxicity is not expected to have specific developmental effects. High dosages of a test substance during the earliest stages could potentially lead to death of the embryo, but at lower doses the embryo could survive with no change greater than a delay in the overall developmental schedule.

As soon as groups of cells are comitted to specific roles in organ formation, they develop more specialized metabolic requirements which presumably result in more specific sensitivities to a test substance's toxicity. Exposure during organogenesis, when cell groups and tissues segregate into organ primordia, is more likely to cause structural alterations. Later in organogenesis "critical periods" of sensitivity are known to occur in some organs and systems, but major structural alterations are less likely to be induced by a test substance after completion of basic organ formation.

Before gross organ structure is completed, finer development occurs at the cellular level (histogenesis) and functional capabilities begin developing. Histogenesis and functional development continue into the subsequent growth phase of most organs, and completion of functional development usually coincides with completion of histological differentiation. Effects of test substances during this stage of development are usually determined by microscopic examination of affected organs

Table 4: End Points of Developmental Toxicity Usually Encountered in Submitted Studies*

Endpoint	Description
	Litters with implants (Number per litter)
Implantation sites	= M
Corpora lutea	= CL; important when treatment begins prior to implantation; is difficult in mice.
Live offspring[1]	= P; offspring refers to fetuses or pups depending on study protocol.
Resorptions[1,2]	= r
Late fetal deaths[1]	= D
Postimplantation loss[1]	= r + D = $M_{nonlive}$
Altered offspring[1,3]	= A; those with external, visceral or skeletal alterations; includes separate counts from each litter and for each type of change.
Affected implants[1]	= A + $M_{nonlive}$
Live offspring[1,4]	= P; measured at selected intervals until termination in postnatal studies; numbers of live males and females are used to determine sex ratio for each litter.
Stillbirths[1]	= S; from postnatal studies

* Adapted From USEPA, 1991; Table 2.

1 The number of litters with a count of one or more for these observations should also be noted, and the percentage of litters affected per number of litters with implants should be considered in a review.

2 The number of litters that are totally resorbed should also be considered.

3 The incidence of malformations and variations should be available in the study report for individual offspring and grouped according to litter and dose

4 The number of live male and female offspring should be reported so that the sex ratio for each individual litter can be determined.

Table 4: (continued)

Endpoint	Description
	Percentages calculated from each litter with implants
Pre-implantation loss	= [(CL - M)/CL] X 100
Live offspring	= [P/M] X 100
Resorptions	= [r/M] X 100
Late fetal deaths	= [D/M] X 100
Nonlive implants	= [$M_{nonlive}$/M] X 100
Affected implants	= [($M_{nonlive}$ + A)/M] X 100
Stillbirths	= [S/M] X 100
	Percentages calculated from each litter with live offspring
Live offspring	= [P/(P + D)] X 100 or [P/(P + S)] X 100 for postnatal studies
Viability	If P_0 is the number alive at the initial observation time and P_t is the number alive at observation t, then % viability at time t is equal to [P_t/P_0] X 100.
Altered offspring	= [A/P] X 100
	Other observations from each litter with live offspring
Body weight	Mean weight of offspring per litter at selected intervals in postnatal studies and at termination of prenatal studies.
Clinical signs	Type, incidence, duration and degree (from postnatal studies)
Gross necropsy and histopathology	(Offspring from postnatal studies)

or postnatal evaluations of affected functions (e.g., behavior, reflexes, motor activity, etc.).

Developmental toxicity can manifest itself as altered growth, structural alterations, functional deficiencies, and death. These effects increase in degree as dosage increases from the no-observed-effect level (NOEL) to the totally lethal level, and different no-effect levels might exist for each type of developmental end point when caused by the same test substance. Some developmental end points may influence the incidence of others. For example, an increase in early embryolethality might preclude the incidence of structural alterations, growth retardation, or the incidence of postnatal functional deficits by reducing the number of offspring at risk. The NOEL should be characterized by the absence of all four types of developmental effects. Table 4 indicates the way in which developmental end points may be expressed.

1. Death

Prior to implantation and before differentiation begins, the early embryo has great regenerative capability (totipotency), but if the dosage is sufficiently high, death of the conceptus may prevent pregnancy or reduce litter size. After implantation and during early stages of organogenesis, susceptible embryos may die and be resorbed. Resorptions can be complete leaving a very small amount of embryonic tissue that is detectable only by staining of the apparently nongravid uterus with ammonium sulfide. In gravid uteri, an implantation scar at the site of implantation (metrial gland) may contain sufficient embryonic tissue to be observed grossly. Some investigators classify these sites as **early resorptions.** As organogenesis progresses, toxic insults to the embryo may still result in resorption. These sites are referred to as **late resorptions** and are indicated by the presence of both fetal tissue and placental tissue at the implantation site. After the period of major organogenesis, the fetus is increasingly resistant to lethality, but dead fetuses (as indicated by their lack of movement) can be observed. The development of major malformations may cause fetal death, so consideration of developmental alterations in dead fetuses should be mentioned in reviews of studies with compound-related fetal deaths.

Control litters are known to have some fetal wastage because of resorptions or fetal death. Therefore, dose-related fetal loss should result in reduced numbers of viable offspring in each litter such that group mean litter sizes in a given treatment group (live fetuses per litter) are statistically significantly less than the control group mean.

In order to properly assess intrauterine death, fetuses should be delivered by caesarian section prior to their expected birth to prevent loss of prenatal material.

Such losses frequently occur in many species after birth since maternal animals often cannibalize stillborn and abnormal pups. Furthermore, evidence of resorptions can only be observed by internal examination of the maternal reproductive tract.

The Agency provides the following guidance on pre- and post-implantation loss (USEPA, 1991b):

> When treatment of females begins prior to implantation, an increase in preimplantation loss could indicate an adverse effect on gamete transport, the fertilization process, uterine toxicity, the developing blastocyst, or on the process of implantation itself. If treatment begins around the time of implantation, an increase in preimplantation loss probably reflects variability that is not treatment-related in the animals being tested, but the data should be examined carefully to determine if there is a dose-response relationship. If preimplantation loss is related to dose, further studies would be necessary to determine the mechanism and extent of such effects.
>
> The number and percent of live offspring per litter, based on all litters, may include litters that have no live implants. The number and percent of late resorptions and fetal deaths give some indication of when the conceptus died, and the number and percent of nonlive implants per litter (post implantation loss) is a combination of these two measures. Expression of the data as the number and percent of litters showing an increased incidence for these end points may be less useful than incidence per litter because, in the former case, a litter is counted whether one or all implants were resorbed, dead, or nonlive.

A complete analysis of postimplantation losses should also consider the number of live offspring per litter. The Agency's risk assessment guidelines explain this point as follows:

> The number of live offspring per litter based on those litters that have one or more live offspring may be unchanged even though the incidence of nonlive in all litters is increased. This could occur either because of an increase in the number of litters with no live offspring or an increase in the number of implants per litter. A decrease in the number of live offspring per litter is generally accompanied by an increase in the incidence of nonlive implants per litter unless the implant numbers differ in all dose groups.

2. Altered Growth

In addition to known genetic, endocrine, and nutritional factors that influence growth, many agents which are capable of producing death or malformations may also cause altered growth. Two parameters often employed to assess altered growth are fetal weight and fetal length (crown-rump length). One may be altered without the other and each is considered to be a developmental toxicity end points.

Developmental Toxicity End Points

Fetal body weight is generally inversely proportional to litter size (i.e., higher fetal weight is usually expected from a smaller litter size and lower fetal weight is associated with larger litter size). Therefore, decreased fetal weight with unaffected or reduced litter size is generally regarded as an indication of developmental toxicity. However, the Agency's risk assessment guidelines (USEPA, 1991b) state that because of the inverse relationship between litter size and fetal weight, "...(expected fetal weight decreases at) the upper end of the dose-response curve may be affected by smaller litters and increased fetal or neonatal weight."

Another factor to consider in the analysis of fetal weights is that males generally weigh more than females in the most commonly used laboratory animal species (USEPA, 1991b). Thus a litter that is predominantly male should tend to have a higher average fetal weight than the litter of mostly females.

Criteria sometimes used to describe offspring as runts (growth-retarded) may include body weight that is two standard deviations less than the group mean fetal weight or fetal weights that fall below a range defined by historical control data. The criterion used by a given investigator should be presented along with the data when a dose-related effect is noted.

Altered growth can be a transient or a permanent effect. In the case of a possible transient effect (reduced fetal weight, delayed ossification, etc.), normal maturation and increases in weight and size will be expected after birth. Whether this recovery after birth represents a complete reversal of all toxic effects on the offspring is unknown. On the other hand, failure to recover from growth retardation is readily accepted as a defect (permanent stunting). The reversibility of growth retardation (including runting) must be assessed by considering appropriate data from conventional developmental toxicity, post-natal, and multi-generation reproduction studies. Reproduction studies are required for all food-use pesticides and may be useful for determining the reversibility of some aspects of fetotoxicity, such as dilated ureters and renal pelvis.

Although effects on fetal sex ratio are quite rare, such data should always be examined since chemical agents may preferentially affect a particular sex (e.g., acetazolamide, Scott et al., 1972). With hormonally active test substances, normal protocols may fail to detect adverse effects on structure and/or function in sexual development unless dosing is continued to the end of gestation since the sexual organs and characteristics are among the last to develop.

3. Structural Alterations

The fetus is usually examined for external, soft tissue, and skeletal alterations. These changes are usually classified as either variations or malformations. **Variations** are generally regarded as alterations that may not adversely affect the fetuses and have no fatal outcome. **Malformations**, on the other hand, are abnormalities that are considered to have a significant adverse effect on the fetus with or without fatal consequence. Distinctions between variations and malformations are sometimes problematic in developmental toxicity studies, and no standard criteria for distinction between the two types of alterations have been universally accepted. A glossary of terms for commonly used developmental alterations is presented in the Appendix of this SEP and examples are illustrated in Figures 1-9 on pages ? through 43.

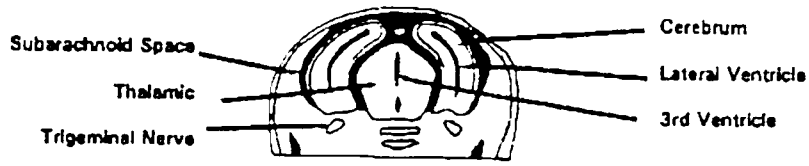
a. Methods for examination of fetuses

All fetuses should be examined for external alterations which are detected by means of a dissecting microscope. Limbs, tail, thorax and abdomen, face, palate, eyes, and head are systematically examined.

The Guidelines (§83-3) indicate that for rodents, approximately one-half to two-thirds of fetuses from each litter should be prepared and examined for skeletal abnormalities and one-third to one-half for soft tissue abnormalities using appropriate methods. For rabbits, "each fetus should be examined by careful dissection for visceral abnormalities and then examined for skeletal abnormalities". It should be understood that despite the Guidelines reference to "visceral", the intention was that soft tissue examination data for both the head and torso be provided. Deviations from the recommended protocol must be explained by the registrant.

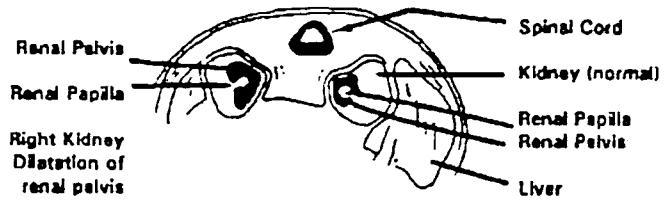
The selected fetuses are examined by the method described by Staples (1977) and Stuckhardt and Poppe (1984) or fixed and examined for visceral abnormalities by free-hand razor blade sectioning (Wilson, 1965). This sectioning technique is adequate for rodent and rabbit fetuses (Barrow and Taylor, 1969. J. Morph. 127:291-306).

A modified version of Dawson's technique (1926) with Alizarin Red S has been used in staining the skeleton for evaluation of alterations. However, one of several techniques may be used depending on the test facility. If cartilage abnormalities are suspected, counter staining techniques using toluidine blue or alcian blue are helpful to detect cartilage in the absence of ossification (Kimmel, et al. 1981 Stain Technol. 56:271-273).



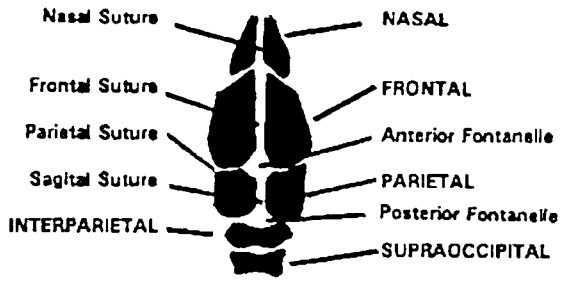
NORMAL

CROSS SECTION THROUGH CEREBRAL HEMISPHERE



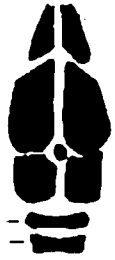
CROSS SECTION THROUGH MID ABDOMEN

Figure 1: Examples of alterations of the limbs and digits



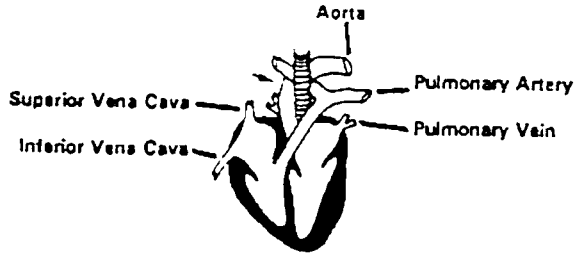
NORMAL

ENLARGED ANTERIOR FONTANELLE
ENLARGED SAGITAL SUTURE



INCOMPLETE OSSIFICATION OF
INTERPARIETAL AND
SUPRAOCCIPITAL

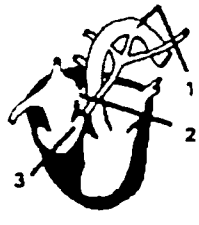
Figure 2: Examples of soft tissue observations (brain and abdomen)



RINGED AORTA

TRANSPOSITION
OF BLOOD VESSELS
(Aorta on right side and
pulmonary artery on left)

TRILOCULAR
(No separation
between ventricles)



1. Patent Ductus Arteriosus
2. Patent Oval Foramen
3. Pulmonary Valvular Atresia

REVERSED AORTA

Figure 3(a): Examples of cardiovascular alterations

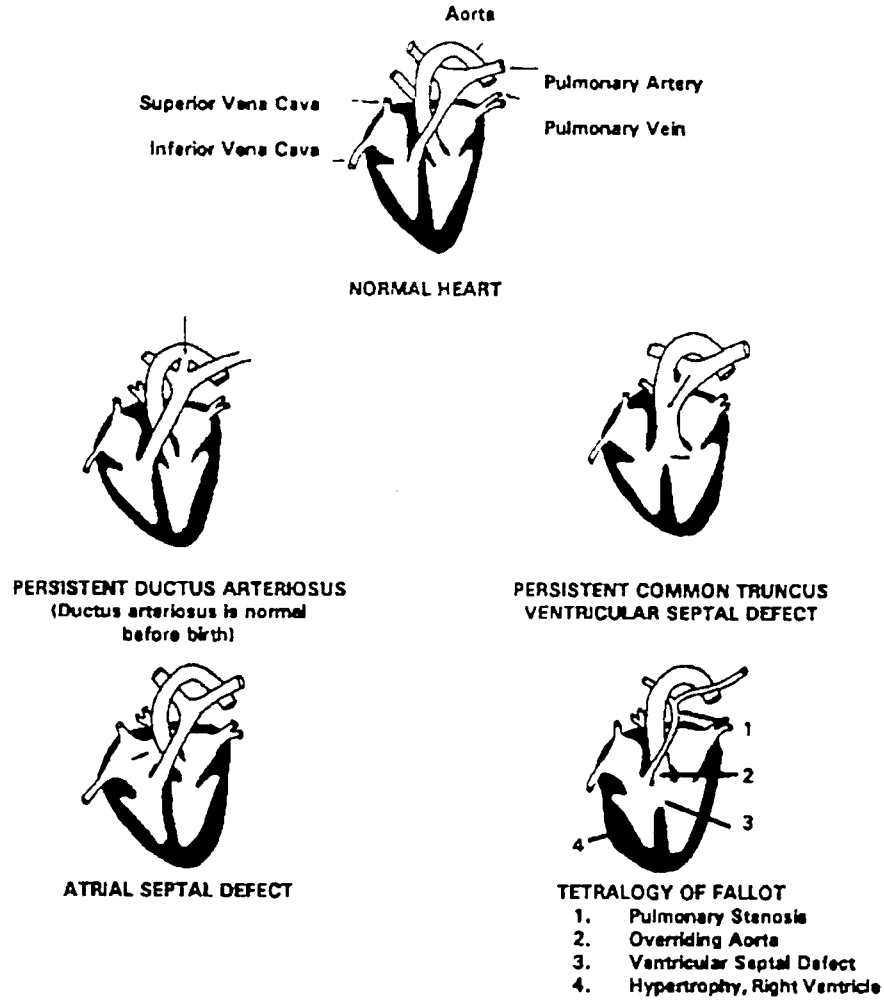


Figure 3(b): Examples of cardiovascular alterations

14th THORACIC CENTRUM

MISSING THROACIC ARCH #9
INCOMPLETELY OSSIFIED
LUMBAR ARCHES

MISSING LUMBAR ARCHES

INCOMPLETELY OSSIFIED
METATARSALS

Figure 4: Examples of urinary tract alterations

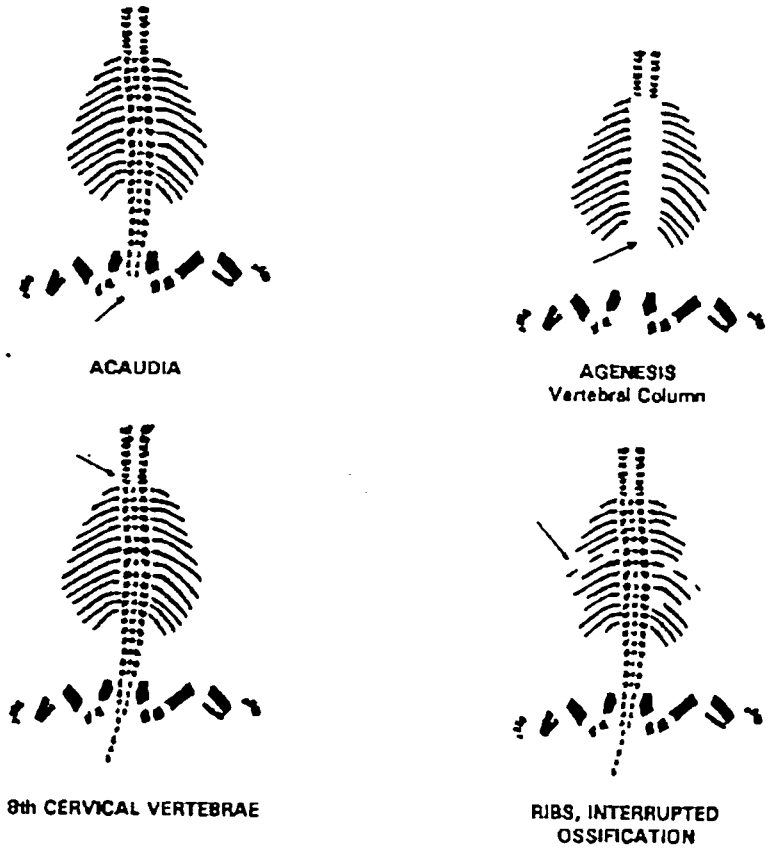


Figure 5: Examples of alterations of skull bone ossification

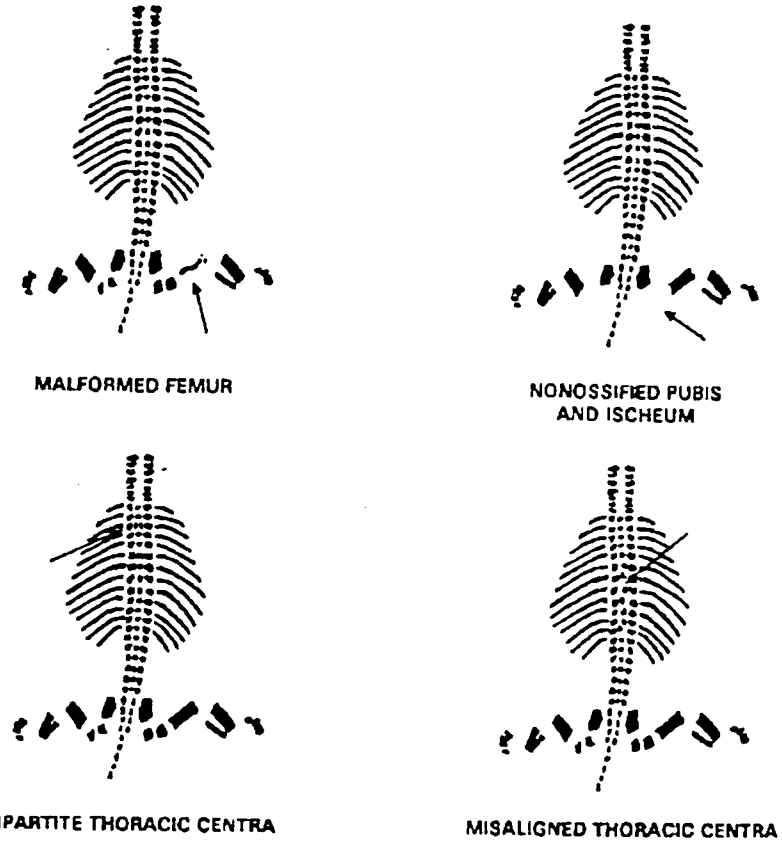


Figure 6(a): Examples of alterations of sternebrae ossification

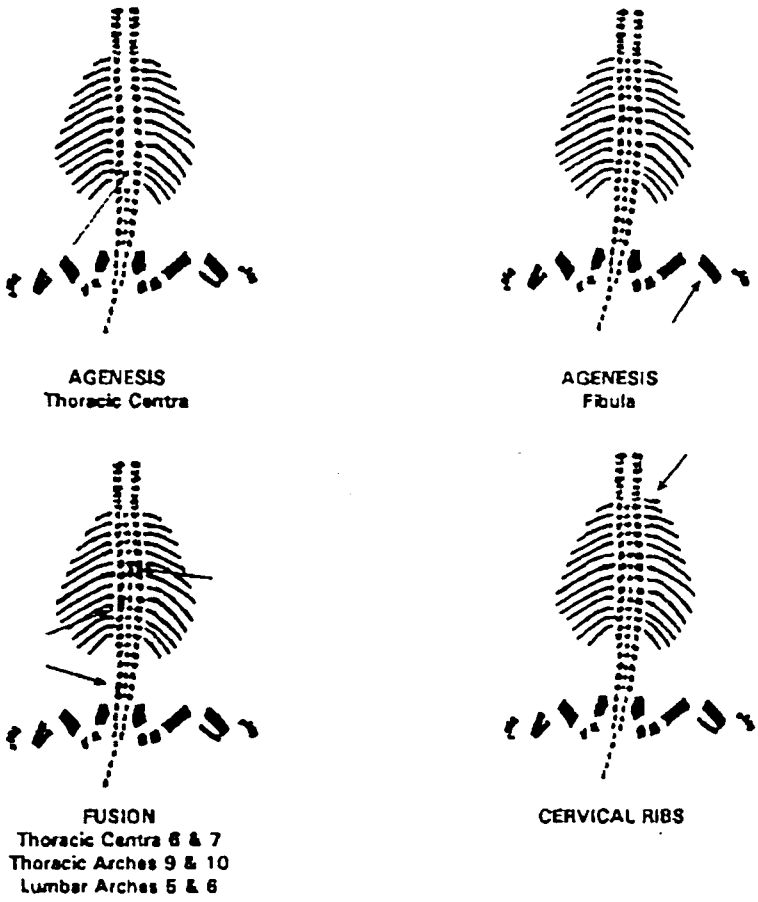


Figure 6(b): Examples of alterations of sternebrae ossification

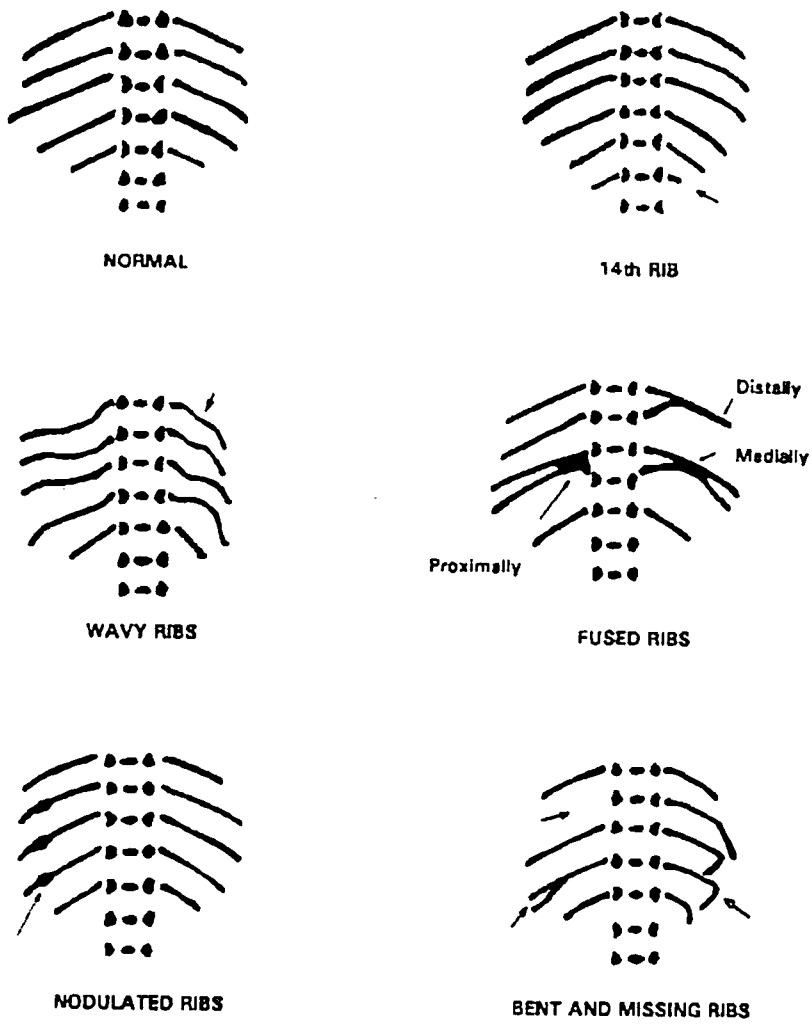


Figure 7(a): Examples of vertebral alterations

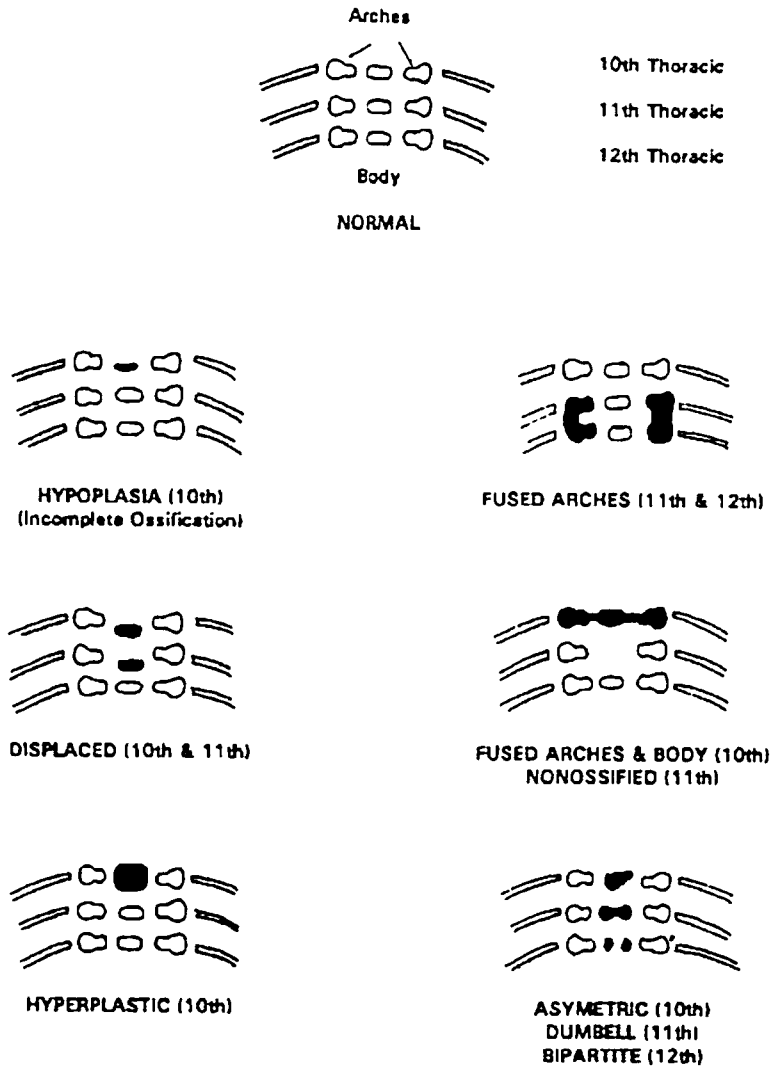


Figure 7(b): Examples of vertebral and other skeletal alterations

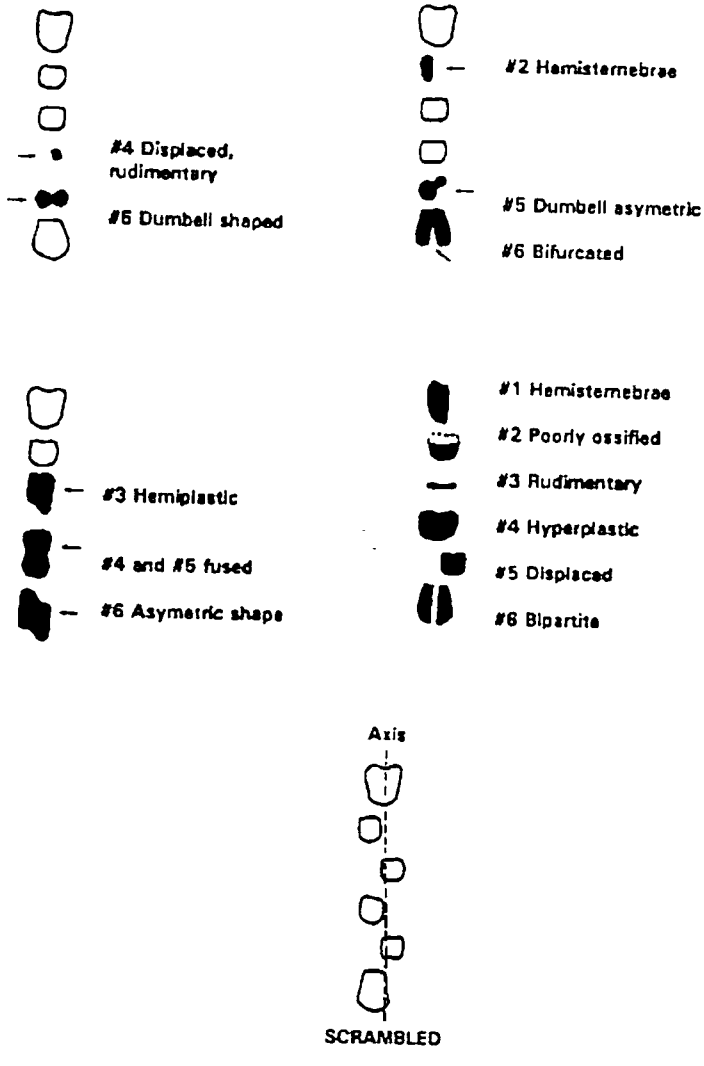


Figure 8: Examples of rib alterations

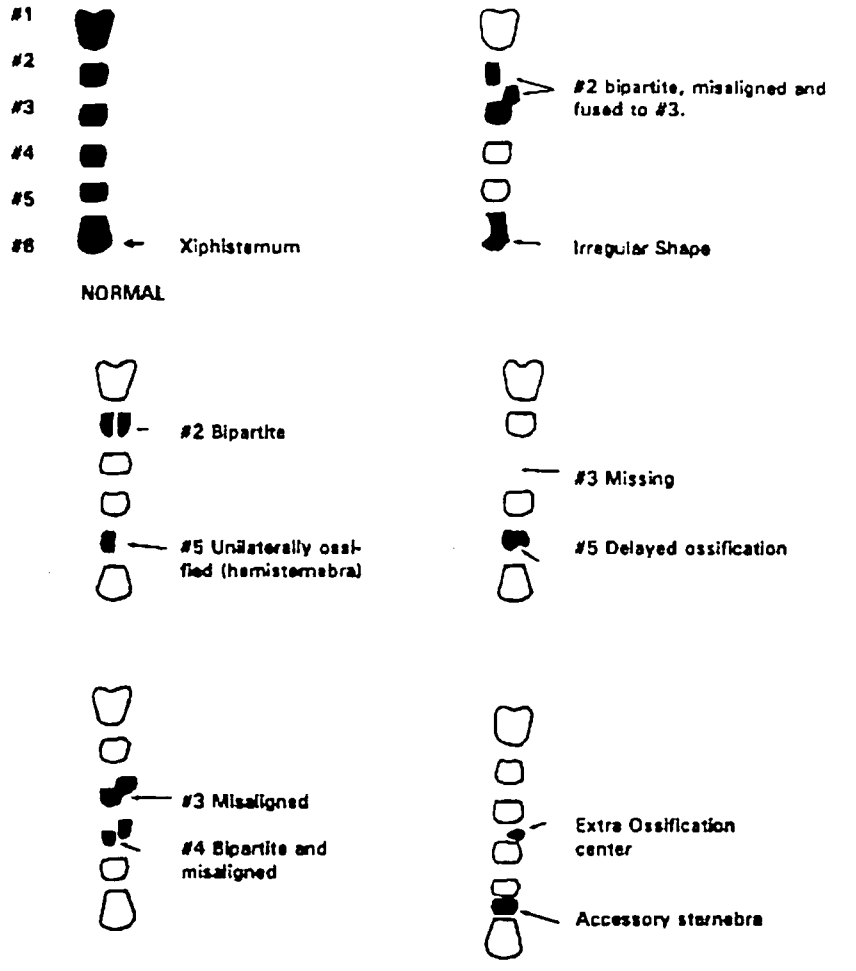


Figure 9(a): Examples of skeletal alterations

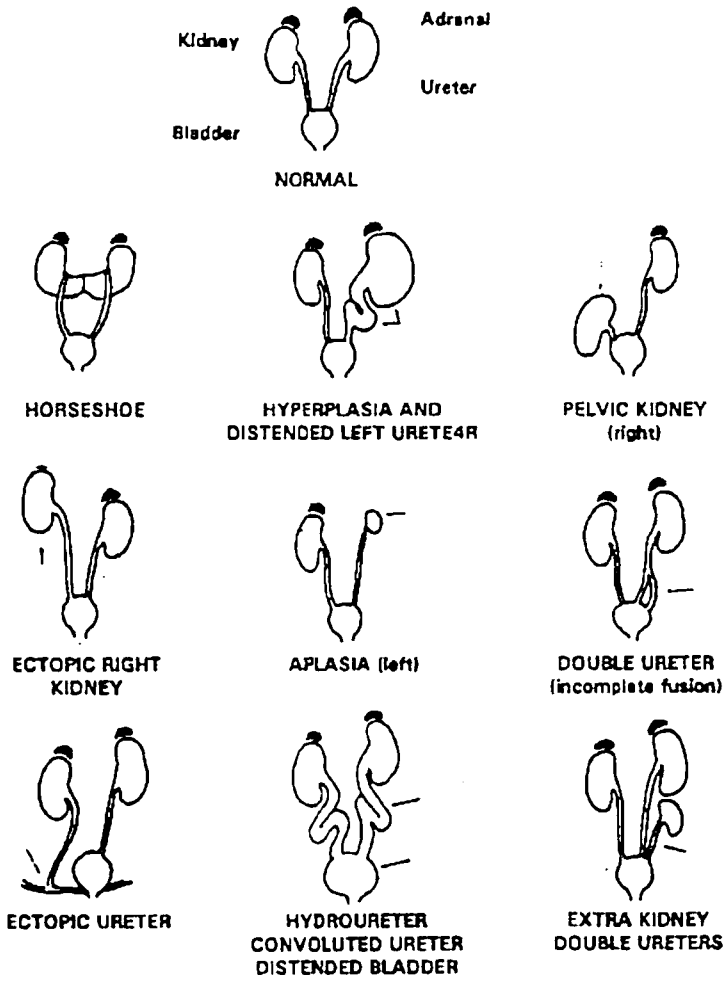


Figure 9(b): Examples of skeletal alterations

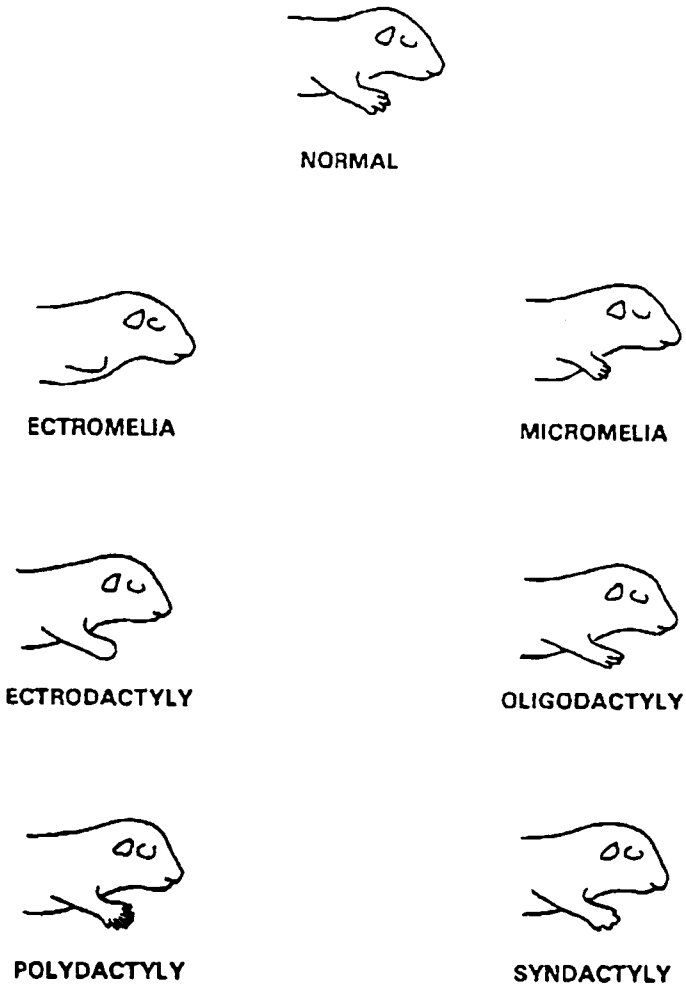


Figure 9(c): Examples of skeletal alterations

b. Interpretation of Results

The risk assessment guidelines (USEPA, 1991b) describe several aspects of examining and interpreting results from developmental toxicity studies as follows:

> A listing of individual offspring with their malformations and variations may give an indication of the pattern of the developmental deviations...The incidence of individual types of malformations and variations may indicate significant changes that are masked if the data on all malformations and/or variations are pooled.
>
> ..., the biological significance of an altered incidence of anatomical variations is more difficult to assess (than a dose-related increase of malformations) and must take into account what is known about developmental stage (e.g., with skeletal ossification), background incidence of certain variations (e.g., 12 or 13 pairs of ribs in rabbits), or other strain- or species-specific factors. However, if variations are significantly increased in a dose-related manner, these should also be evaluated as a possible indication of developmental toxicity.
>
> In addition, some investigators have considered certain of these effects to simply be associated with manifestations of maternal toxicity noted at similar dose levels (Khera, 1984, 1985, 1987), such effects are still toxic manifestations and as such are generally considered a reasonable basis for Agency regulation and/or risk assessment. On a somewhat similar note, the conclusion of the participants in a "Workshop on Reproductive Toxicity Risk Assessment" (Kimmel *et al.*, 1988) was that dose-related increases in effects that may occur spontaneously are as relevant as dose-related increases in any other developmental toxicity end point.

Developmental effects can be localized in one particularly sensitive animal's litter (all conceptuses affected) perhaps because of combined maternal and test substance factors. However, developmental toxicity may also appear in one or two fetuses from several litters in a treated group in which maternal factors may have less influence on fetal response to a test substance. Although the number of affected fetuses may be the same for two groups each with one of these distributions, the interpretation of each type of result should be different. Since the maternal animal, not the conceptus, is directly exposed to the test substance, the number of affected litters should be considered first. Therefore, the distribution of one or two affected fetuses in several litters is of greater concern than the single litter with all fetuses affected because there may be other conditions associated with the developmental effects localized in one entire litter.

The incidence of developmental alterations should be compared to concurrent and historical control data collected from various studies with the same species, strain of animals and vehicle. Since there may be genetic drift in the strain of animals used, a comparison must be made with historical control data limited to a specific time-frame (see next section beginning on page 53).

Developmental Toxicity End Points

Another aspect of the dose-response relationship that may be encountered in developmental toxicity studies is that some developmental end points may influence the incidence of others such that a dose-related decrease in the incidence of an effect may be observed (see Section III. D., page 59). According to the Agency's risk assessment guidelines (USEPA, 1991b), the end point "affected implants," which is the combination of postimplantation losses and malformed conceptuses, may sometimes reflect a dose-response better than each of its two component end points taken separately. The risk assessment guidelines further state,

> This is especially true at the high end of the dose-response curve in cases when the incidence of nonlive implants per litter is greatly increased. In such cases, the malformation rate may appear to decrease because only unaffected offspring have survived. If the incidence of prenatal deaths or malformations is unchanged, then the incidence of affected implants will not provide any additional dose-response information...

For each variation, both the number of fetuses and litters affected are important in making a final conclusion. Also, the pattern of variations should be considered. An increase in the incidence of variations can be assumed to be treatment related if the increase is statistically significant over the control range and particularly if the increase is dose-dependent. Trends should also be carefully considered.

For malformations it should be recognized (Palmer, 1977):

- That almost without exception every type of malformation ever recorded can occur sporadically in any species, and
- That almost every type of malformation can arise from more than one cause.

Based on these two points, an individual occurrence of an extremely rare malformation in the absence of a dose-response may be spontaneous in origin. Conversely, if a larger population (N) were sampled in a repeat study, the possibility also exists that a treatment-related effect may be demonstrated. Finally, Wilson (1977) notes, "It has become axiomatic in experimental teratology that agents capable of causing any adverse biological effects can usually also be shown to be embryotoxic under the right conditions of dose, developmental stage, and species susceptibility, unless maternal toxicity intercedes."

4. Functional Alterations

Functional capabilities begin developing during organogenesis and continue into the subsequent growth phase of most organs. At birth the nervous, reproductive and endocrine systems are not fully functional and metabolic and immunological competence have not fully developed. Therefore, functional effects of test

substances are usually determined by postnatal, multigeneration reproduction, or other non-guideline studies that are not routinely submitted to support pesticide registration. Evaluation procedures for these studies are beyond the scope of this SEP.

However, the Agency has published guidelines for pesticides that are potentially neurotoxic and will need to be evaluated for potential effects on the structure and function of the nervous system in offspring (U.S. EPA, 1991c). In these studies, functional end points may include observations to detect alterations of motor, sensory, learning, and reflex activities such as those listed in Table 5.

Table 5: Examples of Postnatal Parameters Measuring Neonatal Functional Development in the Nervous System.

Reflex Activities	Motor Activities
Airdrop righting	Ambulation time
Cliff avoidance	Ascending vertical rod
Negative geotaxis	Ascending wire mesh
Pain reflex reaction	Parallel bars
Surface righting	Rotating rod
	Rotating drum
	Swimming test
Sensory Activities	**Learning Activities**
Auditory startle	Bar press - avoidance test
Olfactory discrimination	Mazes (Y or T)
Visual placement	Open field

The *Guidelines for Developmental Toxicity Risk Assessment* (U.S. EPA, 1991b) indicate that functional effects resulting from exposure of offspring to a test substance may be observed at dose levels that are lower than those at which other developmental toxicity end points are altered. The guidelines further state, "Such effects may be transient or reversible in nature but generally are considered adverse effects." The Agency further notes (U.S. EPA, 1991b):

> Data from postnatal studies, when available, are considered very useful for further assessment of the relative importance and severity of findings in the fetus and neonate. Often, the long-term consequences of adverse developmental outcomes noted at birth are unknown, and further data on postnatal development and function are necessary to determine the full spectrum of potential developmental effects. Useful data can also be derived from well-conducted multigeneration studies, although the dose levels used in those studies may be much lower than in studies with shorter-term exposure.

> Less work has been done on other developing functional systems, but the assessment of postnatal renal morphological and functional development may serve as a model for the use of postnatal evaluations in the risk assessment process. As an example, standard morphological analyses of the kidneys of fetal rodents have detected treatment-related changes in the relative growth of the renal papilla versus the renal cortex, an effect considered in some cases to be a malformation (hydronephrosis), while in other cases a variation (apparent hydronephrosis, enlarged or dilated renal pelvis). While some investigators (Woo and Hoar, 1972) have providing data suggesting that the morphological effect represents a transient developmental delay, others have shown that it can persist well into postnatal life and that physiological function is compromised in affected individuals (Kavlock et al., 1987a, 1988; Datson et al., 1988; Couture, 1990). Thus the biologicalinterpretation of this effect on the basis of fetal examinations alone is tenuous (U.S. EPA, 1985b). In addition, the critical period for inducing renal morphological abnormalities extends into the postnatal period (Couture, 1990), and studies on perinatally-induced renal growth retardation (Kavlock et al., 1986, 1987b; Slotkin et al., 1988; Gray et al., 1989; Gray and Kavlock, 1991) have shown that renal function is generally altered in such conditions but that manifestation of the dysfunction is not readily is not readily predictable. Thus both morphological and functional assessment of the kidneys after birth can provide useful and complementary information on the persistence and biological significance of expressions of developmental toxicity.

According to the risk assessment guidelines (U.S. EPA, 1991b), the cardiovascular, immune, endocrine, reproductive, respiratory, and digestive systems are susceptible to altered development of competence after exposure to a test substance. The guidelines further state, "Currently there are no standard testing procedures for these functional systems..." Therefore, when such data are available, they should be referred to an appropriately experienced individual before they are considered in the risk assessment process.

5. Maternal and Developmental Toxicity

In the interpretation of developmental toxicity studies both maternal and developmental end points should be considered together. Developmental toxicity is of greater concern when it is observed at doses below maternally toxic levels, but the developmental effects are more frequently reported at maternally toxic dose levels. In the more frequent case, the Agency risk assessment guidelines state, "...the developmental effects are still considered to represent developmental toxicity and should not be discounted as being secondary to maternal toxicity." The guidelines further note that there is currently no basis for the assumption that developmental effects only result from maternal toxicity, and maternal toxicity at a given dose level may be reversible whereas developmental end points may be permanently altered at that level.

C. Reporting of End Points

There are a variety of ways to report results from §83-3 studies, but the differences are specific and relate to how individual animal data are presented and summarized. As mentioned in the introductory discussion of Section II. page 7, studies have been rejected because appendices containing individual animal data (on both maternal and developmental end points) and summary tables of that data were incomplete or unavailable for review, or there was insufficient background information (e.g., results from range-finding studies or historical control data) to aid in characterizing adequacy of test doses or defining NOELs or LOELs.

As a minimum, the Acceptance Criteria (see Section II. A., page 8) and §83-3 test guidelines indicate that a report should contain individual animal and summary data on the following:

- daily observations.
- body weights.
- food consumption (only when the test substance is administered in the diet).
- number of fetal deaths,
- early and late resorptions
- numbers of viable fetuses per sex
- number of corpora lutea (except when the test species is the mouse).
- litter weights and/or fetal weights/sex/litter.
- litter and fetal incidences of external, skeletal, and soft tissue malformations and variations

Tables 3 and 4 (see pages 20 and 24, respectively) indicate that the incidences of *in utero* and fetal observations can be reported as the number or percentage per litter. Numbers and percentages for viable fetuses (live offspring) are reported for litters with implants and litters with live offspring (i.e., excluding totally resorbed or aborted litters). Results are also summarized in reports as the number of litters with a given effect per number of litters examined (i.e, the number of litters containing one or more early resorptions, fetal deaths, fetuses with a specific alteration, etc.), and some reports also include total fetal incidences for effects (i.e., the total number of fetuses in a group regardless of litter with a specific alteration per total number of fetuses examined for that effect). Since the litter is the experimental unit, Tables 3 and 4 list end points that are always expressed as a count or proportion per litter in a group, and data based on total fetuses regardless of their litter of origin should generally not be considered.

A sample data set is shown in Tables 6, 7, and 8 to illustrate an approach to the variety of reporting formats submitted for evaluation. The data were taken from an acceptable study in rabbits. Since the individual data came from the highest dose group and since that dose level was excessively toxic, the example is not typically

acceptable data. The data set was chosen because relationships between end points are apparent and important to any interpretation of developmental toxicity data.

The group initially contained 16 animals, and one of those was nonpregnant. Nine were sacrificed or died before the end of the study (five aborted and four died during treatment). Table 6 shows the individual animal data for maternal body weight and how the maternal losses affected the number of animals available for calculations of group mean body weights, especially late in gestation (see section III. E., page 56). The data from Table 6. were also used in the original report to calculate body weight changes for specific periods during gestation. For example, the body weight change for animal #6 during gestation days 7-13 is 4173 g minus 4370 g = -197 g, which contributed to the group mean change of -127 g for the entire group (the control group showed a mean change of -9 g for gestation days 7 through 13). Table 6 indicates that all 16 animals were used to determine the body weight change for gestation days 7-13, but later post-dosing intervals used fewer animals (15 on day 20, 13 on day 24, and 6 on day 29).

Table 6 also illustrates the use of adjusted maternal body weight (the reported group mean for this end point was 3693 g compared with a control group mean of 4231 g), but this end point could be derived from only six animals (see Section III. A. 2. above).

Of the six litters available for evaluation in this data set, all had pre- and postimplantation losses. Individual animal results of uterine and fetal observations are presented in Tables 7 and 8, and observed values for Animal #6 are summarized with reported high dose and control group means in Table 9. The results shown in Table 9 demonstrate that submitted reports may not include summary data for every end point listed in Table 4 (page 24). In this example, postimplantation losses are not clearly broken down to early and late resorptions and fetal deaths. However, Table 7 shows that these losses were observed in 4, 2 and 2 of the 6 litters, respectively. The missing information can usually be obtained from individual animal data if it is necessary to support an interpretation of the study's results, but additioal time is needed to complete this type of detailed review.

Similar analysis of individual animal data from the other groups in the study could be conducted if the reported postimplantation losses indicated an effect. In this case the individual animal data for the control group indicates that 3 of 12 litters had one early resorption (one was in a doe with only one implant), 2 of 11 litters (excluding the totally resorbed litter) had one late resorption each, and 1 of 11 control group litters had a dead fetus. This pattern is similar to that of the high dose group in that most of the postimplantation losses are early resorptions in both groups. There were results in the other groups of the study that showed no increase in postimplantation

Table 6: Sample Individual Maternal Body Weights (grams) (a dose group from a rabbit study)

Animal number	Gestation Day						Uterus weight (g)	Adjusted[2] body weight (g)
	0	7	13	20	24	29		
1	4168	4024	3806	3362	Died on gestation day 21			
2[1]	3800	3469	3708	2823	2708	2466	-[4]	-[4]
3	4108	4152	4101	3763	3538[3]	Aborted on gestation day 24		
4	4758	5036	4806	4306	4162	Died gestation day 27 (aborted prior to death)		
5	4454	4474	4320	3936	3788	Died gestation day 26		
6	4204	4370	4173	4240	4276	4308	389.9	3918.1
7	4604	4798	4544	4096	3942	Died on gestation day 26		
8	4264	4370	4064	3600	Aborted on gestation day 23			
9	4262	4468	4344	3874	3632	3450	167.9	3282.1
10	4400	4926	4924	4706	4552	4394	276.7	4117.3
11	4018	4124	4084	3666	3514	Aborted on gestation day 26		
12	4876	4962	5002	4501	4300	4306	226.8	4079.2
13	4748	4923	4940	4657	4370	4136	245.3	3890.7
14	4552	4970	4394	Died on gestation day 19				
15	3912	3974	3838	3336	3178	2978	107.3	2870.7
16	5068	5216	4802	4435	4320	Aborted on gestation day 25		

[1] Nongravid, not included in calculation of mean.
[2] Doe body weight on gestation day 29 minus gravid uterus weight..
[3] Body weight recorded after abortion, not included in calculation of the mean.
[4] Not applicable, animal was nongravid.

Table 7: Sample Individual Maternal and Fetal Observations Made at Caesarean Section in a Developmental Toxicity Study with Rabbits.

Animal number	Number of: Corpora lutea	Implantations	Resorptions: Early	Resorptions: Late	Dead fetuses	Post implantation loss	Live fetuses	Sex distribution: Male	Sex distribution: Female	Mean fetal body weight (g)
1	Died on gestation day 21 (gravid)									
2	Nongravid									
3	Aborted on gestation day 24									
4	Died on gestation day 27 (gravid), aborted prior to death									
5	Died on gestation day 26 (gravid)									
6	11	7	1	0	0	1	6	2	4	46.3
7	Died on gestation day 26 (gravid)									
8	Aborted on gestation day 23									
9	8	6	1	1	1	3	3	3[1]	1	17.1[2]
10	13	7	0	2	0	2	5	3	2	28.7
11	Aborted on gestation day 26									
12	15	5	0	0	2	2	3	4[1]	1[1]	24.9[2]
13	13	6	1	0	0	1	5	2	3	31.5
14	Died on gestation day 19 (gravid)									
15	12	5	2	0	0	2	3	1	2	21.9
16	Aborted on gestation day 25									

[1] Includes a dead fetus.
[2] Excludes dead fetuses.

Table 8: Sample of Individual Fetal Variations Observed in a Rabbit Developmental Toxicity Study.

Animal number	Number of fetuses examined			Numbe3r of fetuses with variations	Fetus number	Description of variation
	Externally	Viscerally	Skeletally			
6	6	6	6	5	5	Left carotid arises from inominate
					5, 7	27 presacral vertebrae
					1	13th rudimentary rib
					3, 5, 6, 7	> 12 pairs of full ribs
9	4	4	4	4	1	Azygous lobe of lung absent
					3	Hyoid body unossified
					1	27 presacral vertebrae
					1, 2	> 12 pairs of full ribs
					1, 2, 3, 4[1]	Sternebra(e) #5 and/or #6 unossified
					2, 3	Pubic bone unossified
					2	Tail unossified
10	5	5	5	3	7	Left carotid arises from inominate
					2	13th rudimentary rib
					1, 7	Sternebra(e) #5 and/or #6 unossified
12	5	5	5	5	1	Gallbladder smaller than normal
					1, 2[1]	27 presacral vertebrae
					1, 3[1], 5	> 12 pairs of ribs
					2[1], 4	Sternebrae #5 or #6 unossified
13	5	5	5	4	4	13th rudimentary rib
					1, 3	> 12 pairs of ribs
					6	Sternebra #5 unossified
15	3	3	3	3	3	Azygous lobe of lung absent
					1, 4	Hyoid body unossified
					1	27 presacral vertebrae
					4	13th rudimentary rib
					1, 3	> 12 pairs of ribs
					1,4	Sternebra #5 unossified
Total	28	28	28	25	-	

[1] Dead fetuse.

loss in the presence of lesser maternal toxicity. (Analysis of the other results is byond the scope of this discussion.)

Table 9: Individual animal data from Animal #6 in Tables 6 and 7 compared with reported for the animal's dose group and control group means.

End Point	Animal #6	Dose Group Mean	Control Group Mean
Corpora lutea per doe	11	12	15.2
Implantations per doe	7	6	7.2
Postimplantation loss per doe	1	1.8	0.5
% Preimplantation loss per litter	36.4	50.0	49.1
% Postimplantation loss per litter	14.3	30.6	7.1
Live offspring per litter			
with implants	6	4.2	6.7
with viable fetuses	6	4.2	6.7
Sex distribution			
total number of males	2	15	38
total number of females	4	13	43
Resorptions per litter			
Early	1	- [a]	- [a]
Late	0	- [a]	- [a]
Late fetal deaths	0	- [a]	- [a]
Nonlive implants per litter [b]	1	1.8	0.5
Altered fetuses [c]			
total per litter	5	- [a]	- [a]
with >12 pairs of full ribs	4	13 (5) [d]	1 (1) [d]

[a] These values were not included in summary tables, but their absence is not critical to the acceptability of the study since the dose level was associated with excessive toxicity, and results from lower dose groups did not show a test substance related effect on early and late resorptions or fetal deaths. These values can be determined from the individual animal data.

[b] This end point was referred to as "Postimplantation loss", and there was no separate listing for early and late resorptions or fetal deaths. As indicated in the previous footnote, this detail of data reporting is not critical to the interpretation of results. These details can be determined from individual animal data.

[c] Includes viable and nonviable fetuses (see discussion on page).

[d] Number of altered fetuses (number of litters containing one or more altered fetuses).

D. Using Historical Control Data

A true concurrent control is one that is the same as treatment groups except for the administration of the compound being evaluated. All experimental procedures should be duplicated in control groups just as they are in treated groups. Historical control data is a compilation of results from control groups that are concurrent to a group of studies conducted at the same laboratory under the same experimental

conditions. Similarly, Historical control data appropriate to a study under review should be generated by the same laboratory using the same experimental procedures followed in the study being evaluated. The historical data should be supplied from the test laboratory within at least a two year range and should be presented by individual study with appropriate descriptive statistics for the sample of control groups (e.g., means from mean values for each control group in the data base, the standard deviation of the historical control mean, the range of control group means observed in the historical data base, etc).

Because of personnel changes, slight differences in experimental procedures that may not be reported, or subtle changes in the laboratory environment over time, historical control data should not be used as a substitute for concurrent control results in an analysis of developmental toxicity end points (Kimmel and Price, 1990 as cited in USEPA, 1991b).

Historical control data can provide a guide for determining the biological significance of statistically significant differences observed in a developmental toxicity study. Such data may indicate whether an alteration induced by a test compound is rare, uncommon, or common in untreated animals, or historical data can identify a concurrent control group incidence that may be unusually low for the test species.

An unusually low incidence of an effect in a concurrent control group may result in statistically significantly increased incidences in dosed groups. These apparent increases may not be unusual for similar sized groups of untreated animals of the strain and species tested. The mean and standard deviation of the mean for the sample of historical control groups are more appropriate than the range in this type of analysis because the ends of the range may themselves be unusual.

Historical control data may also indicate trends in the incidence of spontaneously occurring alterations. An uncommonly observed alteration may become more common over time because of a suspected genetic drift in the test species population at a laboratory or because of an improved technique of observation.

Circumstances such as a slightly increased incidence of an effect or examination of a small number of litters in test groups (e.g., results from a range-tinding study) preclude the detection of statistically significant differences, the incidence of developmental effects in a dose group should be compared with historical control data. If the dose group has an incidence of an effect that is outside the historical range, if the concurrent control group's incidence of the same effect is not below the historical range (i.e., unusually low), and if other dose levels show a dose-related increase in the effect, then a significant effect can be assumed. Otherwise, the historical control data can indicate that the apparent increase in a developmental

alteration is within normal ranges for the species and strain tested, the concurrent control group did not have a representative incidence of effect being considered.

Historical control data were appended to the study from which the sample data set described in the previous section was taken. In that collection of 24 studies the mean number of postimplantation losses per litter was reported to be 0.8 with a range from 0.3 to 1.7. The distribution of those results is as follows:

Postimplantations per litter	Number of studies
0.3	1
0.4	1
0.5	5
0.7	4
0.8	3
0.9	3
1.0	1
1.1	3
1.2	2
1.7	1

The number of postimplantation losses per doe in the high dose group of the sample data set was 1.8 which is just outside the historical range. The control group in the sample data averaged 0.5 postimplantation losses per litter which is a commonly observed incidence.

E. Statistical Analysis

The design of most teratology studies submitted for evaluation follow a standard guideline (§83-3), and the statistical methods are consistent from study to study with occasional exceptions. This section provides a limited overview of the methods routinely applied and characteristics of results that may be encountered in a study evaluation. More rigorous discussions of the principles and methods described here can be found in statistics references such as Gad and Weil (1986 and 1982), Gaylor (1978), Sokal and Rolf (1981), and Snedecor and Cochran (1989). Table 10 summarizes the end points and some of the most commonly used methods for analysis of developmental toxicity data. Other statistical methods not listed in Table 10 are often used, and the list included here should not be considered complete. An experienced statistician should always be consulted when there are doubts about statistical methods described in reports under review.

Statistical Analysis

The most important aspect of developmental toxicity data analysis is that the litter is considered the experimental unit. Effects on a treated female and her litter are biologically independent of effects that may occur in other females and their litters in a study. Effects of a test substance on each fetus in a litter are related to the status of the treated animal bearing that fetus. Because of individual differences in maternal susceptibility, an entire litter can be affected, while others in the same dose group can be unaffected. This means all fetuses in a single dose group are not equally at risk to the potential developmental effects of the test substance. Therefore, the accepted practice is to consider the litter as the experimental unit for developmental toxicity studies (Gad and Weil, 1982; Gaylor, 1978; etc.).

Sometimes individual animals are excluded or censored from consideration because they may be inappropriate to an analysis of results for a given end point. For example, investigators exclude animals in the analysis of body weight gains during gestation because the animals were not pregnant, or group means for resorptions will exclude litters that are totally resorbed from group mean resorption rates. Aborted pregnancies or animals that died pregnant are considered separately from those in the group that survived to the end of the study (see Tables 6 and 7 on pages 50 and 51. These losses are anticipated by using groups containing more animals than the number of litters recommended for examination at the end of the study. This practice minimizes the effect of losses on the study's capacity to detect significant group differences that may be caused by the test substance.

To assure that erroneous conclusions are not reached for certain types of data (see Table 10), investigators sometimes test for homogeneity of variances using Bartlett's test. If the test shows variances to be heterogeneous then adjustments may be made as follows:

- outliers are removed and Bartlett's test is repeated on the data;
- data are transformed and reevaluated with Bartlett's test; or
- if neither of the two previous procedures equalized variances, a distribution-free statistical method such as the Kruskal-Wallis test or the Wilcoxon-Mann-Whitney U test is used in hypothesis testing.

In comparing two or more groups with respect to the proportion of litters with dead, resorbed, or altered fetuses, there should be no significant differences in the average number of implants per litter for each group. If the number of implants is not significantly different, then the probability that each litter can be affected is roughly similar, and an effect on the incidence of litters with dead, resorbed, or altered fetuses could be detected. After implantation data have been analyzed for significant

differences and none are found, the number of litters in each group containing one or more altered, resorbed or dead fetuses can be compared using the tests suggested in Table 10.

The incidence of fetuses with some abnormalities may also increase the percentages of dead fetuses per litter, and a statistical analysis of the percentage of normal live fetuses per litter may be more appropriate in an evaluation of the significance of those effects in general. Some abnormal fetuses may be resorbed or have a higher death rate, and the percentage of abnormal fetuses per litter may not appear to be unusual when calculated as a percentage of live fetuses only. The percentage of live fetuses per litter would be expected to decrease significantly if a test substance increased the incidence of lethal fetal alterations. Therefore, the percentage of live fetuses per litter should be a part of the statistical evaluation of a study's results.

The power of a study is defined as the probability that it will demonstrate a true effect (U.S. EPA, 1991a). A study's power is limited by the following factors:

the sample size used in the study,

the background incidence of the end point under consideration,

the variability in the incidence of the end point, and

the analysis method.

The risk assessment guidelines provide the following example to illustrate how these factors affect a study's ability to detect group differences:

> ...the number of litters needed to detect a 5% or 10% change was dramatically lower for fetal weight (a continuous variable with low variability) than for resorptions (a binomial response with high variability). With the current recommendations in (§83-3)..., the minimum change detectable is an increased incidence of malformations is 5 to 12 times above control levels, an increase 3 to 6 times the in utero death rate, and a decrease 0.15 to 0.25 times in the fetal weight.

Statistical methods are also used to characterize the relationship between an independent variable such as dose levels and dependent variables such as end points of developmental toxicity (model fitting). These methods are occasionally described in developmental toxicity studies received in the Health Effects Division and the Agency is encouraging the use of model fitting techniques to determine the benchmark dose (see Section IV. C. 68), but they will not be discussed in detail here.

Table 10: Data Characteristics and Commonly Used Statistical Methods for Hypothesis Testing in Developmental Toxicity Studies (adapted from Gad and Weil, 1986)

End Points	Commonly Used Statistical Tests
Body weights, body weight change, food consumption, organ weights (absolute & relative), survival rates,[1] and crown-rump length	Bartlett's test for homogeneity of variances,[2] F test[3] Analysis of variance (ANOVA),[4] t test[4]
Behavioral signs (some), corpora lutea, implantation sites, and live and dead fetuses	Wilcoxon-Mann-Whitney U test,[6] Kruskal-Wallis test[6]
Behavioral signs, clinical signs, fetal alterations,[5] dose/ mortality data	Chi square test,[7] Fisher's Exact test

[1] End points are expressed as proportions or percentages of each litter with pre- or post-implantation losses, early or late resorptions, and live or dead fetuses.

[2] When Bartlett's test indicates that variances are not homogeneous, data are sometimes transformed by using the square root, the arcsin of the square root or other transformations of each value to stabilize variances before further analyses are attempted. Because some of the statistical tests have become standard practice, Bartlett's test is not always used.

[3] The F test is the same type of test as Bartlett's except that it tests for homogeneity of variances for just two groups.

[4] ANOVA is used for comparison of three or more groups of data with homogeneous variances and a normal frequency distribution. The t test compares two groups of continuous normally distributed data.

[5] Group incidences (number of litters with affected fetuses/number examined in each group) are compared.

[6] If $n \geq 40$, a 2 by 2 chi square test with continuity correction can be done.

[7] For $n \leq 9$ nonparametric methods should be used because normality and homogeneity of variance are decreased. The Wilcoxon-Mann-Whitney U and Kruskal-Wallis Tests are nonparametric tests that make no mathematical assumptions about the distribution of the data. If $n \geq 10$, a t test is just as useful.

Model fitting may include such methods as regression analysis, Jonckheere's test, andother methods which are beyond the scope of this Standard Evaluation Procedure. Questions regarding this aspect of statistical analysis in developmental toxicity studies should be referred to a statistician.

F. Evaluation of Dose-Response in a Single Study

Ideally, a dose-response should identify a NOEL and an LOEL. The highest dose level at which there is no statistically or biologically significant increase in the frequency of an adverse effect on developmental end points when compared with the appropriate control group is the NOAEL. The LOAEL is the lowest dose at which there is a significant increase in adverse effects on developmental end points.

As mentioned in the introductory discussion in Section III., the manifestations of developmental toxicity increase in frequency and degree with increasing dosage from the NOEL to the totally lethal level. Previous discussions also indicated that maternal factors and relationships between certain developmental end points affect a dose-response for one particular end point. Because of these factors, results can represent part or all of a dose-response curve, and there are several possible patterns of response that may be encountered. These patterns are:

(1) an increase only at the highest dose tested,

(2) a NOAEL and two higher doses with increasing responses (an ideal situation),

(3) an increase for all treated groups that correlates with the increase in dose levels,

(4) an increase for all treated groups that correlates inversely with the increase in dose levels,

(5) an increase that is similar for all treated groups at all dose levels,

(6) an increase at the lowest dose group only,

(7) an increase at a mid dose group only, and

If the NOAEL and LOAEL are not clear in a study, other dose-response information from range-finding studies, historical control data, or a second study in the same test animal should be evaluated to identify the two values for the test species.

Evaluation of Dose-Response in a Single Study

The risk assessment guidelines (U.S. EPA, 1991b) note that focusing on the NOEL does not incorporate information on the slope of the dose-response curve or the variability in the data. Because data variability is not considered, the guidelines state, "...the NOAEL will likely be higher with decreasing sample size or poorer study conduct, either of which is usually associated with increasing variability in the data." Range-finding studies are usually done with groups that are one-quarter to half the size of those used in a principal study and are not as powerful in detecting developmental effects. In addition, range-finding studies are designed to evaluate the high end of a dose-response curve as a reference for selecting doses in principal studies which are designed to investigate lower portions of the curve. Therefore, a range-finding study should be used in a qualitative way in the evaluation of a dose-response.

For example, near the high end of the dose-response curve when nonlive implants per litter may be greatly increased, the malformation rate in live offspring may actually decrease with increasing dose because only unaffected implants have survived. The end point "affected implants," which is the combination of postimplantation losses and malformed conceptuses, may sometimes reflect a dose-response better than each of its two component end points taken separately. However, data from a range-finding study are variable, and they are generated for a purpose other than to identify NOAELs or LOAELs. They should only be used to estimate the range of the dose-response curve evaluated by an associated principal study.

Considering end points such as "affected implants" may be useful in explaining dose-response patterns (4), (6), and (7). Another explanation for such patterns of dose-response may be the correlation between increased corpora lutea which leads to increases in the number of pre- or postimplantation losses without affecting the number of live fetuses per litter. Combining the incidences of types of individual malformations or variations may mask significant changes that are indicated by one type which could also lead to dose-response patterns (4), (6), and (7).

As noted in the discussion beginning on page 53, historical control data may indicate whether the incidence of an alteration in a treated or control group is within the range of incidences observed in untreated animals. An unusually low incidence of a developmental alteration in a concurrent control group may result in an apparent increased incidence in a dose group which may not be unusual for similar sized groups of untreated animals of the strain and species tested. Comparisons of results representing dose-response patterns (1) and (5) with historical control data can be used to support conclusions about the significance of the observed increases. If the increased incidences in the treated groups are above historical the historical range, they should be considered evidence of a dose-response for (1) and a treatment-related

effect for (5). For dose-response patterns such as (3), the historical control data may be used to establish a NOAEL.

IV. DEVELOPMENTAL TOXICITY RISK ASSESSMENT

As mentioned in Section I. B. 8., the three parts of a developmental toxicity risk assessment are hazard identification/dose response assessment, exposure assessment and risk characterization. This section discusses the use of developmental toxicity study reviews in those three parts of the risk assessment process for pesticides that cause developmental toxicity in laboratory animals. Since human evidence is referred to qualified scientists for evaluation and is rarely available on pesticides, the discussions that follow will focus on laboratory animal data.

A. Hazard Identification/Dose Response Assessment

After each developmental toxicity study on a given pesticide has been accepted and reviewed, the risk assessment guidelines recommend that the data should collectively be classified as "Sufficient Evidence" or "Insufficient Evidence." "Sufficient Evidence" includes the subcategories of "Sufficient Human Evidence" and "Sufficient Experimental Animal Evidence/Limited Human Evidence." The risk assessment guidelines provide the following descriptions for the two data categories considered in the sections that follow:

> The sufficient evidence category includes data that provides enough information to judge whether or not a human developmental hazard could exist within the context of dose, duration, timing and route of exposure. This category includes both human and experimental animal data.
>
> The minimum evidence necessary to judge that a potential hazard exists generally would be data demonstrating an adverse developmental effect in a single, appropriate, well-conducted study in a single experimental animal species. The minimum evidence needed to judge that a potential hazard does not exist would include data from appropriate well-conducted laboratory animal studies in several species (at least two) which evaluated a variety of the potential manifestations of developmental toxicity, and showed no developmental effects at doses that were maternally toxic.
>
> (Insufficient evidence)...includes situations for which there is less than the minimum evidence necessary for assessing the potential for developmental toxicity, such as when no data are available on developmental toxicity, as well as for data bases from studies in animals or humans, that have a limited study design (e.g., small numbers, inappropriate dose selection/exposure information, other uncontrolled factors) or data from a single species reported to have no adverse developmental effects, or data bases limited to information on structure/activity relationships, short-term tests, pharmacokinetics, or metabolic precursers.

There are five assumptions about should be considered in evaluating a data base for the potential of a test substance to cause developmental toxicity (U.S. EPA, 1991b). These assumptions are:

- An agent that produces an adverse developmental effect in experimental animal studies will potentially pose a hazard to humans following sufficient exposure during development.
- All of the four manifestations of developmental toxicity (death, structural abnormalities, growth alterations and functional deficits) are of concern.
- The types of developmental effects seen in animal studies are not necessarily the same as those that may be produced in humans.
- In the absence of data to the contrary (e.g., pharmacokinetics or mechanisms of action), humans are as sensitive or more so than the most sensitive animal species tested.
- A threshold exists for the dose-response curve of test substances that produce developmental toxicity.

Evaluation of individual developmental toxicity studies should provide information on the following:

- quality of the data (see Section II., page 7),
- the sensitivity of the studies (see Section III. E., page 55),
- the number and types of end points affected (Tables 3 and 4, pages 20 and 24),
- adequacy of doses tested (see Section II. B. 2. a., page 10),
- route of administration (see Section II. B. 3., page 14)

Other characteristics of the pesticide's toxicity that are considered include:

- pharmacokinetic data,
- structure-activity relationships, and
- other toxicological data (e.g., reproduction toxicity, mutagenicity, carcinogenicity, etc.).

1. Using multiple studies in hazard identification

Typically, there are from one to multiple developmental toxicity studies available on any given pesticide. Factors to be considered when using all available studies for hazard identification are as follows:

- reproducibility of results,
- the number of species affected,

When multiple studies are available for risk assessment, the data base as a whole should be classified into one of the categories of evidence described above. When all the studies comprising the data base do not meet testing guidelines, scientific

judgement should be used to determine whether additional testing is needed or whether the studies can be used together in satisfying the criteria for "Sufficient Evidence." For example, in a group of studies that do not meet current testing guidelines, there may be two studies which provide conflicting results (i.e., one suggests a potential for developmental toxicity and the other does not). If these studies are comparable with respect to the route, level, and duration of exposure, another test may be required in an attempt to see which of the two studies' results can be reproduced. However, if the two studies in this example were conducted at different dose ranges and the study evaluating the higher range suggested developmental toxicity, results from both tests may be combined to determine a NOAEL and an LOAEL.

For pesticides, the most common data base consists of two standard guideline §83-3 studies using three dose groups and a control group. As noted in Section III. D., above, the evaluation of dose-response relationships in each of these studies attempts to identify effect levels as well as doses that are associated with no increased incidence of adverse effects when compared with controls (the NOAEL). Those adverse effects observed at the lowest dose level (the LOAEL), which may be any of the four manifestations of developmental toxicity, are the critical effects. The NOAEL should be the highest level of exposure under the conditions of the animal study that is not associated with a significant increase in adverse effects. The critical effect from the most sensitive of the two test species is used for determining the NOAEL or LOAEL in deriving a reference dose or concentration for developmental toxicity (RfD_{DT} for oral or dermal exposure, and RfC_{DT} for inhalation exposures).

The risk assessment guidelines (U.S. EPA, 1991b) define the RfD_{DT} and RfC_{DT} as:

> ...an estimate of a daily exposure to the human population that is assumed to be without appreciable risk of deleterious developmental effects... The RfD_{DT} or RfC_{DT} is derived by applying uncertainty factors to the NOAEL (or the LOAEL if an NOAEL is not available), or the benchmark dose...

Uncertainty factors (UFs) applied to the developmental and maternal NOAELs include a 10-fold factor for interspecies variation and a 10-fold factor for intrasepcies variation (UF = 100). When only an LOAEL is available, uncertainty factors up to 10 may be applied (UF $\leq$ 1000), depending on the sensitivity of the end points evaluated, the dose levels tested, or general confidence in the LOAEL. Other modifying factors (MFs) that can be applied to the NOAELs include variability within species, the slope of the dose-response curve, background incidence of the effects, the route of administration, and pharmacokinetic data.

The total uncertainty factor selected is divided into the NOAEL or LOAEL for the critical effect in the most appropriate and/or sensitive study to determine the RfD_{DT} or RfC_{DT}.

Jelovsek, et al. (1990) have suggested some "rules of thumb" which represent an approach used by many experts in developmental toxicology to assess results from multiple studies. Some of these principles are summarized as follows:

- A well designed study (one that follows the principles discussed in Sections II. and III.) that shows a pesticide has developmental toxicity has more weight in a hazard assessment than a poorly designed study showing no developmental effects.

- When a study using larger numbers of animals per group (>25 for example) contradicts a study in which fewer animals are used (<12 for example), the larger study is given more weight.

- In animal studies, consistency in the pattern of developmental effects noted in several experiments (replication of results) has more significance than several reports of inconsistent patterns of affected developmental toxicity end points.

- A study showing no developmental effects in which there was no maternal toxicity is considered to be less sensitive than a negative study that demonstrated maternal toxicity (see Section II. A. 1. b. i.).

However, these "rules of thumb" should be used with the five assumptions presented in the discussion on page 62 in mind. For example, a poorly conducted study suggesting developmental effects that are not observed in a well-conducted study should be interpreted as weak evidence of a potential human hazard.

2. Using other information

a. Reproduction studies

Data from single or multigeneration studies can be useful in confirming findings from teratology (§83-3) studies, or they may detect developmental effects (e.g., functional alterations) that are unlikely to be induced by exposure during just the period of major organogenesis. Continued exposure of offspring from conception through maturation in reproduction studies provides more opportunities to assess the potential for developmental toxicity over most of the development of offspring. Although a detailed discussion of the use of reproduction studies in risk assessment is beyond the scope of this document, it is reasonable to evaluate the need for additional developmental studies based on effects reported in reproduction studies.

b. Pharmacokinetic and physiological considerations

Pharmacokinetic information on a test substance can be helpful in the assessment of developmental toxicity data from more than one test species as well as in extrapolation between species. The risk assessment guidelines point out:

> Information on absorption, half-life, steady-state and/or peak plasma concentrations, placental metabolism and transfer, excretion in breast milk, comparative metabolism, and concentrations of the parent compound and metabolites may be useful in predicting risk for developmental toxicity. Such data may be helpful in defining the dose-response curve, developing a more accurate comparison of species sensitivity, determining dosimetry at target sites, and comparing pharmacokinetic profiles for various dosing regimens or routes of exposure. Pharmacokinetic studies in developmental toxicology are most useful if conducted in animals at the stage when developmental insults occur. The correlation of pharmacokinetic parameters and developmental toxicity data may be useful in determining the contribution of specific pharmacokinetic parameters to the effects observed.

i. Differences between pregnant and non-pregnant animals

Physiological changes in several organ systems during pregnancy can alter the pharmacokinetics of a pesticide in the test species (Mattison, 1991). These changes are required for successful pregnancy and lactation, and they maintain maternal homeostatic mechanisms for delivery of essential nutrients to fetuses and removal of heat, carbon dioxide and waste products from fetuses. The physiological alterations are species dependent (e.g., cardiac output increases by 50% in humans and by 20% in rabbits), and may involve different physiological strategies. A summary of some of these changes is presented in Table 11.

ii. Differences between laboratory animals and humans

Nau (1991) has noted the following differences between humans and animals with respect to pharmacokinetic and physiological considerations:

- the half-life of xenobiotics (drugs and toxic agents) is an order of magnitude shorter in animals than in humans;
- during conventional developmental toxicity studies, high and sharp concentration-time peaks are often produced because of high clearance rates; and
- these high peaks rapidly fall to insignificant levels.

Humans are exposed to more persistent levels of xenobiotics because of the following factors:

- the half-life of xenobiotics is longer than it is in animals;
- the first pass effect is often, but not always, much more extensive in animals than in humans, and humans may detoxify xenobiotics by different pathways;

- maternal plasma protein binding is often more extensive in humans than it is in animals; and
- the duration of sensitive developmental processes in animals is often several-fold shorter than those of humans (see Table 12).

Table 11: Physiological Changes During Pregnancy*

Parameter	Change
Absorption	
Gastric emptying time	Increased
Intestinal motility	Decreased
Pulmonary function	Increased
Cardiac output	Increased
Blood flow to skin	Increased
Distribution	
Plasma volume	Increased
Total body water	Increased
Plasma proteins	Decreased
Body fat	Increased
Metabolism	
Hepatic metabolism	±
Extrahepatic metabolism	±
Plasma proteins	Decreased

* Taken from Mattison et al., 1991, Table 1

B. Exposure Assessment

Developmental toxicity risk assessments include estimates of occupational exposure (mixer/loader applicators, pilots, etc), dietary and drinking water exposures and residential exposure. Occupational and residential exposure estimates are provided by the Occupational and Residential Exposure Branch of HED while dietary exposure estimates are generated by the Dietary Exposure Section of the Science Analysis Branch (SAB) of HED. It is the responsibility of the Toxicology Branches of HED to adjust occupational and residential estimates of dermal exposure according to dermal absorption studies when they are available. If dermal absorption data are not available, 100% dermal absorption is assumed for purposes of risk characterization until such data become available. For characterization of a dermal risk, direct comparison of the exposure estimate with a NOEL from a dermal developmental toxicity study may also be done.

Table 12: Developmental Rate (days) in the Rat and Human.

Organ/System	Rat	Human
Central Nervous System:		
Neural Fold Closing	10.0	21
Three Brain Vesicles	10.5	26
Cerebral Hemisphere	12.0	29
Olfactory Bulb	13.5	47
Eyes:		
Optic Vesicle	10.5	23
Lens	11.5	32
Optic Nerve Fibers	14.0	48
Eyelids Close	18.0	70
Eyelids Open	38.0*	140
Ear:		
Optic Vesicle	10.5	22
Cochlear Duct	12.7	44
Cardiovascular:		
S-Shaped Heart	10	22
Aortic Arches	10-13	21-37
Septation Begins	11.5	28
Urinary System:		
Pronephros	11.0	21-27
Mesonephros	12.0	27-37
Metanephros	12.8	35-37
Reproductive System:		
Sex of Gonad apparent	14.5	45-48
Degeneration of Mullerian Duct completed in Males	19-22	85
Degeneration of Wolffian Duct completed in Females	19-21	100
Skeletal Systems:		
Upper Limb Appearance	11.0	28-85
Lower Limb Appearance	11.3	31-35
Cartilage (ribs) Appearance	15.0	42-44
Ossification (ribs) Begins	17-18	49

* 16 days postnatally.

C. Risk Characterization

Estimates of daily exposures in units of mg pesticide per kg female body weight are compared with the no-observed-effect level (NOEL) which is also expressed in units of mg/kg/day. The Margin of Exposure (MOE) is calculated as follows:

$$MOE = \frac{NOEL\ (mg/kg/day)}{Exposure\ estimate\ (mg/kg/day)}$$

The Agency has recognized limitations associated with using the NOEL in risk characterization (USEPA, 1991b). These limitations include:

- Dose levels having developmental effects and the slope of the dose-response curve are ignored.
- The variability in the data is not considered.
- Studies failing to establish a NOEL must be repeated, and their results are needlessly discarded.

The ability of a developmental toxicity study to determine the "true" value of the NOEL for a pesticide depends upon the spacing of the doses selected for the study and the power of the study to detect a true difference between treated and untreated animals.

Because of these limitations, the *Developmental Toxicity Risk Assessment Guidelines* (USEPA, 1991) note that the Agency is evaluating calculation of the **benchmark dose** for comparison with the NOEL in risk characterization for developmental toxicants. The benchmark dose is defined for any toxicological end point as lower statistical confidence limit for a dose corresponding to a specified increase in the level of a health effect over the background level (Crump, 1984). Technical aspects of this new method are described elsewhere (Crump, 1984; and Kimmel and Gaylor, 1988). They involve statistical methods of model fitting that are beyond the scope of this Standard Evaluation Procedure, and a more detailed discussion in the risk assessment guidelines.

V. WRITING STUDY REVIEWS AND SUPPORT DOCUMENTS

Once data have been evaluated for acceptability and interpreted using the principles described in previous sections of this Standard Evaluation Procedure, the results must be described and summarized in support documents. The most basic of

these documents is the Data Evaluation Record (Report) commonly referred to as the DER which provides information used in risk assessments. This section discusses the information that should be included in a DER without regard to a specific format or style. In addition, this section provides an outline for a developmental toxicity risk assessment support document based on the one followed in the Agency's presentation of guidelines for developmental toxicity risk assessment (USEPA, 1991b).

A. The Data Evaluation Record

1. Cover Sheet

The DER should be considered a scientific document that is consistent with accepted methods of technical writing (e.g., see "Instructions to Contributors" in *Teratology, Toxicology and Applied Pharmacology, Fundamentals of Applied Toxicology*, etc.).

The first page of the DER should contain the following (see Example below):

- **Identification of HED staff** who conducted the primary and secondary reviews by name, section and branch;
- **Identification of study type** by name (developmental toxicity study) and species tested;
- **Identification of the test chemical** by name (including synonyms), composition, structure (when possible), EPA Pesticide Chemical Code (Shaughnessy number), HED Chemical Number (Caswell number, Tox. Chem. No., etc), EPA Registration Number;
- **EPA'S identification of the study report** by Master Record Identification number (MRID No.), Data Submission Number (numbers given a "D" or "S" prefix on the PRATS sheet accompanying the package), and Health Effects Division project number;
- **Study reference** to include authors, title, testing laboratory, sponsor and/or submitter, sponsor and laboratory numbers for the report, date issued.
- **Summary and conclusion** containing the dose levels tested, the strain of animal used, duration of dosing, route of administration, no-observed-effect levels (NOEL) for maternal and developmental effects, lowest-observed-effect levels (LOEL) with a brief description of the effects and how they changed with dose;
- **Evaluation of study acceptability** which should 1) state whether the study satisfies the §83-3 requirement for registration (see Section I. B.), 2) describe deficiencies in the study and state whether they can be rectified by submission of additional information or conducting another study, and 3) classify the study (assign a "Core classification" of guideline, minimum, supplementary, or invalid, see Section II. E. above for discussion).

Example Cover Sheet for a Data Evaluation Record

Primary Review by:
{title} , Review Section {#}, Toxicology Branch {I or II}/HED
Secondary Review by:
Section Head, Review Section {#}, Toxicology Branch {I or II}/HED

DATA EVALUATION RECORD

Study Type: Teratology - Developmental Toxicity
Guideline §83-3
Species: Rat/Rabbit

EPA Identification No.s:
EPA MRID No.
EPA Pesticide Chemical Code
Toxicology Chemical Code
HED Project No.
Data Submission No.

Test Material: {name used for chemical tested in study, structure should be included.}

Synonyms: {any synonyms (common names, registrant designations, CAS chemical name) provided in the report}

Sponsor: {who requested the study, i.e. the registrant; include address if possible}

Study Number(s): {all numbers used to identify the report}

Testing Facility: {laboratory conducting the study, include address if possible}

Title of Report: {complete title used on the report cover sheet}

Author(s): {all authors of the report}

Report Issued: {date presented on report cover sheet}

Conclusions: {summary of findings, executive summary-put in format intended for the one-liners, include species & strain of animal tested, doses tested, how administered, how many days (gestation days), NOEL's LOEL's, etc.}

Core Classification: {See Section II E.}

This study does/does not satisfy the guideline requirements (§83-3) for a developmental toxicity study in rats/rabbits.

{if study is classified as supplementary data, discuss requirements for a potential upgrade, or if inadequate,

- **Compliance:** indicate whether the report included signed statements of confidentiality, compliance with GLP's, quality assurance, or flagging criteria (FIFRA §6(a)(2) criteria as described in 40 CFR 158.34) were provided.

Much of this information is used to prepare a "Toxicology One-Liner" for a database maintained by the Toxicology Branches in HED. A "One-Liner" for developmental toxicity studies includes the following information from the above list:

Citation	Material	MRID No.	Results	Core grade Document #
Guideline number, Study type, Species, Testing facility, Lab. report number, Date report issued	Active ingredient (%)		DER summary and conclusions	(Document # is assigned after DER is completed.)

2. Materials and Methods

The "Materials and Methods" section of the DER should contain descriptions of the following:

- **Test Animals** (see Sections II. B. 1., page 11): species, strain, supplier, age and body weight at start of study, animal husbandry practices that differ from GLP's (e.g., analysis of food and water if available or if the results have an effect on the outcome of the study).

- **Mating Procedures** (see Section II. B. 1. a., page 12): describe the type (natural or artificial insemination) and techniques used; include male:female ratio, criteria used to determine success of mating (e.g., presence of vaginal plug, presence of sperm in vaginal smears, etc), characteristics of semen used in artificial insemination (e.g., cell density, sperm motility, dilution factors, etc.), hCG used to induce ovulation (source, amount administered, etc).

- **Test Substance** (see Section II. A. 1. a. page 9): purity (% active ingredient), density (if provided), physical description of the material, lot number or batch number, supplier, date of receipt, list of contaminants (If available, this should be included in a Confidential Business Information appendix to the DER).

- **Vehicle** (see discussion on page 14 and Table 2, page 15): include similar information as is needed for the test compound.

- **Dose Solution** (see page 14): frequency of preparation, volume, method for calculating amount of test compound to be used (based on body weight for which gestation day or days), results of analysis for stability and concentration if available, the basis for selection of dose levels (refer to a range-finding study if available or briefly describe range-finding study).

- **Study Design** (see Section I. B. 8. page 5): include dosing schedule (gestation days on which the test compound was administered), dose levels, and group assignment of animals as following:

Test group[1]	Dose level (mg/kg/day)[2]	Number assigned
Control		
Low dose		
Mid dose		
High dose		

[1] If more than one control or middle dose group, indicate separation (e.g., low-mid dose group, high-mid dose group, etc.

[2] ppm if test compound is in the diet; mg/l if test compound is administered by inhalation.

- **Observations** (see Sections III. A. and B. above): descriptions of methods, schedules, and procedures for methods used for end points described in Tables 3 and 4 above; mention use of 10% ammonium sulfide to detect implantation sites in supposedly pregnant animals, Wilson's free-hand razor blade or Staples technique for examining fetuses, staining techniques used such as Dawson's, etc.).

- **Historical Control Data** (see Section III. D., page 53) if provided for comparison with concurrent controls and treated groups, describe number of studies used to compile the historical data base and give a list of specific end points included.

- **Statistical analysis** (see Section III. E., page 55 and Table 10, page 58): mention methods used and comment on acceptability of the procedures.

3. Reported Results

This section of the DER should be divided into two distinct parts. The first should include discussion of the results from observation of maternal end points (see Section

III. A. and Table 3 above) and developmental toxicity end points (see Section III. B., page 23 and Table 4, page 24). It is important to include the number of animals, fetuses, and litters examined in each group along with the descriptive statistics (mean, standard deviation, etc) for each set of significant data since the group size may change according to the end point considered and since statistical significance is not always consistent with biological significance. Data should be summarized in the DER when it is important in defining a NOEL, determining the adequacy of the doses tested, or supporting any conclusions about the developmental toxicity potential of the test material. Statistically significant differences should always be noted with a *p* value and the name of the statistical test used to get it. Calculations of means or indexes that are done independently of the report under review should be identified and methods clearly described.

a. Maternal observations

List all reported mortality, whether animals were found dead or sacrificed moribund and whether they were pregnant or not. Include cause of death if reported, especially when dosing errors are indicated (lung congestion, or fluid and evidence of esophageal trauma). Significant clinical observations can be presented as text or in tabular form (e.g., as number of observation days per number of animals presenting each observation or the number of animals in each group with one or more instances of each sign). The fate of every maternal animal should be accounted for so the number of litters examined can be justified.

Results from observations of group maternal body weight, weight gain and food consumption (if available) should be summarized for the pre-dosing, dosing, post-dosing, and entire dosing periods. A "corrected" body weight gain should also be calculated for the entire gestation period when possible. This value is determined by subtracting the gravid uterine weight from the terminal body weight, and that number is then subtracted from the initial body weight for the animal. These data are important in the DER even when no effects were observed because they are needed to support conclusions that the dose range may not be adequate to induce maternal effects.

b. Developmental observations

Significant results of external, visceral and skeletal examinations of fetuses should be summarized. Data should include the number of pups and litters examined,

the number of pups affected, and the number of litters containing one or more fetuses with each significant effect in each test group. Results for effects that may not be related to treatment should sometimes be included when a study is being used to confirm previous observations or to support a conclusion that the test material has no potential to cause developmental toxicity.

4. Discussion

This section of the DER should contain discussions of findings relative to setting NOEL's and LEL's, differences in interpretations made by the investigators and the reviewer, all study deficiencies and problems, the need for additional information that may be provided by the study sponsor or laboratory, and the basis for Core supplementary or invalid study classifications. It should also be noted that separate NOEL's are determined only for maternal and developmental toxicity, not for embryotoxicity, fetotoxicity, or teratogenicity. The latter three terms are subsets of developmental toxicity, and they are not considered separately when defining a developmental toxicity NOEL.

B. Peer Review Support Document

After completion of the review of developmental toxicity studies on a pesticide and when a hazard is indicated, information from the DER is incorporated into a support document that will be presented to the Health Effects Division's Peer Review Committee for Developmental and Reproductive Effects. An outline for such a document may follow that of the Agency's *Guidelines for Developmental Toxicity Risk Assessment* (USEPA, 1991) as follows:

Suggested Outline for Developmental
Toxicity Peer Review Committee Presentations

I. Introduction

(Includes a brief description of the uses for the chemical and its chemical names, synonyms and structure.)

II. Qualitative Assessment of Relevant Data

A. Rat Study #1

1. Description of maternal toxicity (including data to show dose-response and extent of effects). This should provide the Committee with sufficient information to arrive at their own conclusions regarding the appropriateness of dose selection.

2. Description of developmental toxicity (including data to show dose-response and type of developmental effects). This should provide the Committee with sufficient information to arrive at their own conclusions.

3. Briefly summarize deficiencies and limitations of the study.

B. Rat Study #2

1. (Same as A. 1., above)
2. (Same as A. 2., above)
3. (Same as A. 3., above)

C. Rat Study #n (same as A. and B., above)

D. Rabbit Study (same as rat study summary outline)

III. Discussion of Other Evidence

A. Subchronic and Chronic Toxicity Data

B. Reproduction Studies

C. Mutagenicity Studies

D. Metabolism/pharmacokinetics/physico-chemical data

E. Structure-Activity Relationships

IV. Discussion of Strength of the Evidence

A. Strength of the Evidence

1. the quality of the data,
2. the resolving power of the studies,
3. the number and types of end points examined,
4. the relevance of route and timing of exposure,
5. the appropriateness of dose selection,
6. the reproducibility of the effects,
7. the number of species examined,
8. pharmacokinetic data,
9. structure-activity, and
10. other factors

B. Questions to the Committee

1. What is the most appropriate data set and end points (developmental toxicity, maternal toxicity, etc) to use in the risk assessment?

2. Should the most sensitive species be used?

3. What is an appropriate follow-up (i.e., additional data from registrant, exposure assessment to characterize the potential risk, etc.)

V. Appendixes with supporting data

A minimum of two appendixes should follow the document outlined above. They should contain the DER's for the developmental toxicity and reproduction toxicity studies and the Toxicology "One-Liners". Additional appendices may be needed (e.g., historical control data).

APPENDIX: GLOSSARY

APPENDIX

GLOSSARY OF TERMS

The following glossary of terminology for developmental toxicity was compiled from many sources (Benirschke, Garner, and Jones, 1978; Blood, and Studdert, 1988; *Dorland's Illustrated Medical Dictionary*, 1976; Farris and Griffith, 1967; Hafez, 1970; Mid-Atlantic Regional Teratology Association (MARTA); National Research Council, 1989; *Stedman's Medical Dictionary*, 21st ed, 1966; and Thomas, 1981). The glossary provides information on developmental parameters and definitions for developmental alterations observed in laboratory animals and humans.

Many developmental alterations in animals can be described adequately by analogous terms more appropriate to human developmental changes. Human terminology is often the only description available for an alteration. In this glossary, selection of human terminology to define a developmental alteration in animals may not always be appropriate. For example, the term talipes (club foot) may not be precisely analogous across species, but it may serve as a guide or general description. Therefore, a major portion of this glossary consists of terms appropriate to alterations of human development.

Each entry of the glossary consists of a term which is printed in a bolded and enlarged font. The term is separated from its definition by a hyphan, and synonymns are listed inside square brackets. An asterisk precedes terms taken from the MARTA glossary cited above, and most of the remaining entrys were compiled by Dr. Quang Bui. An entry has the following format:

> **Term** - Definition or relevant information. [syn: list of synonymns]

A

abasia - Inability to walk from a defect in coordination, specifically, an inability to walk inspite of other possible muscular movements and sensations.

abasophalangy - Absence of phalanges most proximal to metacarpals or metatarsals. (Literally means inability to walk phalanges).

abdominal hernia - See omphalocele.

ablepharia - Absence, partial or complete, of eyelids.

abnormality - Any deviation from normal. In a study of developmental toxicity, an abnormality means any dose-related or compound-related deviation from normal, including malformations, structural abnormalities, variations, and other deficit. [syn: anomaly].

abortion - Premature expulsion of the products of conception including non-viable fetus(es). Very rare in rodents, but relatively frequent in rabbits. [Syn: miscarriage (in humans)]

acampsia - Abnormal immobility or consolidation of a joint. Also see ankylosis.

acardia - Absence of heart.

acaudal - Absence of a tail [syn.: anurous, acaudate].

acaudate - Without a tail [syn.: anurous, acaudal].

***accessory finger(s)** - Postaxial accessory finger or skin tag. Also see postaxial polydactyly, polydactyly (Type A), and skin tag.

accessory sternebrae - An extra sternebrae is seen completely ossified between two normal sternebrae.

***accessory thumb(s)** - Preaxial accessory thumb; it differs from postaxial polydactyly with it's accessory finger or skin tag. Also see polydactyly (Type B).

acephaly or acephalia - Absence of a head.

acheiria - Absence of one or both hands.

acheiropodia - Absence of hands and feet.

achondroplasia - General size reduction, limbs disproportionally reduced due to defect in cartilage formation at the epiphyses of long bones. Also called chondroplasia fetalis.

***achondroplastic dwarf** - A type of dwarf, having relatively large, short wide head, saddle nose, short extremities, and usually lordosis. See achondroplasia.

acomia - No hair, or naked skin [syn.: alopecia].

acorea - Absence of the pupil of the eye.

acrania - Partial or complete absence of cranium.

acrocephalia - Pointed head or dome shaped head. [syn: oxycephaly]

***acrocephalosyndactyly** - Pointed head and syndactyly of four extremities. [syn: Apert's disease/syndrome and acrosphenosyndactylia]

acystia - Congenital absence of the bladder.

adactyly - Absence of digits of the hand or foot.

adermia - Absence of skin.

agastria - Absence of a stomach.

agenesis - Absence or imperfect development of any part as in thoracic centra agenesis. Organ developmental failure, frequently used to designate failure of the organ primordium to develop. Also see aplasia.

agenesis of the fibula - Absence or imperfect development of the fibula.

agenesis of the thoracic centra - Absence or imperfect development of one or more thoracic centra.

agenesis of the vertebral column - Absence of the vertebral column. The cervical vertebrae may be present.

aglossia - Absence of tongue (see glossi).

aglossostomia - Absence of tongue and mouth opening.

agnathia - Amandibular, absence or virtual absence of lower jaw.

***agyria** - Abnormal or absence of the convolutions of the cerebral cortex. The brain is usually small. [syn: lissencephaly]

***Albers-Schoenberg disease** - See osteopetrosis.

Albright's disease or syndrome - Manifestations including fibrous dysplasia of multiple bones, large pigmented cutaneous nevi with irregular margins and precocious sexual development from endocrine disturbance. [syn: polyostotic fibrous dysplasia]

Albright-McCune-Sternberg syndrome - Fibrous dysplasia of bone, melanotic pigmentation of the skin, and sexual precocity in the female. [syn: osteitis fibrosa disseminata]

alopecia - See **acomia.**

altered growth - Alteration in the growth of an organ, tissue, or body weight. It may be accompanied by alterations of crown-rump length and/or skeletal ossification. Alterations are frequently reversible, and postnatal studies may detect this apparent reversibility.

amelia - Absence of one or both limbs. Also see phocomelia.

amesophalangy - Absence of intermediate phalanges.

ametria - Absence of the uterus.

ammonium sulfide, 10% aqueous solution - Macroscopic identification of implantation sites (metrial glands) in rodent uteri using 10% aqueous (v/v) ammonium sulfide solution. The procedure does not reveal additional sites, but does make existing sites more distinct. [syn: the Salewski method]

amyelia - Absence of spinal cord.

amyoplasia, congenital - Disorder characterized by multiple congenital contractures (arthrogryposis) and joint deformity with almost total absence of skeletal muscle which is replaced by fatty tissues. See arthrogryposis multiplex congenita.

***amyotrophy** - Atrophy of muscle tissue.

anasarca - Generalized massive edema.

anedeous - Absence of genitalia.

anencephaly - Absence of the cranial vault and almost or total absence of the brain or brain reduced to two small masses attached to base of the skull. [Syn.: anencephalia]

anephrogenesis - Absence of renal tissues.

angiectasis - Abnormal dilatation of blood vessels.

aniridia - Complete or partial absence of iris.

anisomelia - Two paired limbs of different lengths.

***ankyloglossia** - Tongue tie.

ankylosis - immobility or inconsolidation of a joint.

anodontia - Absence of some or all teeth.

anogenital distance - Distance between the genital organ and the anus; i.e., from the papilla to the anus (about 2.8 mm in males and about 1.2 mm in female rats at birth).

anomaly - Any deviation from normal. In a study of developmental toxicity, an anomaly is a general term meaning any dose-related or compound-related deviation from normal, including malformations, structural abnormalities, and variations. [syn.: abnormality].

anonychia - Congenital absence of some or all nails.

anophthalmia or anopia - Absent or vestigial eyes, unilateral or bilateral.

anorchism - Congenital absence of one or both testes.

anostosis - Failure of bones to ossify.

anotia - Congenital absence of the external ear.

antenatal - Period between conception and delivery.

antimongolism syndrome - Partial or total deletion of chromosome 21.

anurous - Absence of tail. [syn: acaudal or acaudate]

***aorticopulmonary fenestration** - *See aortic* septal defect.

***aorticopulmonary septal defect** - See aortic septal defect.

***aorticopulmonary window** - See aortic septal defect.

***aortic septal defect** - Abnormal communication between the ascending aorta and the pulmonary artery above the semilunar valves. [syn: aorticopulmonary fenestration, aorticopulmonary septal defect, aorticopulmonary window]

Apert's disease or syndrome - Congenital malformation consisting of a pointed shape of the top of the head and syndactyly (webbing) of the digits. [syn: acrosphenosyndactylia, acrosphenosyndactyly, acrocephalosyndactylia, acrocephalosyndactyly, and Apert's disease]

aphakia - Absence of eye lens.

aphalangia - Absence of one or more phalanges of the forepaws and/or hindpaws.

aplasia - Failure of the cellular components of organs or tissue to develop. See agenesis and hypoplasia.

aplasia of the kidney - Abnormally small development of the kidney due to fewer cells than normal.

aplasia of the ureter - Failure of the ureters to development.

aprosopia - Absence of part or all of the face.

***arachnodactyly** - Abnormal length and slenderness of fingers and toes. [syn: acromacria, dolichostenomelia and spider finger, Marfan's syndrome]

***arhinencephalia** - *Absence of the* rhinencephalon. Rhinencephalon is considered to be the part of the brain concerned with olfactory function, including the olfactory nerves, bulbs, tracts, and the limbic system (not primarily olfactory in function); it is homologous with the olfactory portions of the brain in lower animals. [syn: arrhinoencephia, olfactory brain, smell-brain, and rhinencephalon]

aristocardia - Displacement of the heart to the right side.

Arnold-Chiari deformity - Anomaly of cerebellum and medulla oblongata, which is elongated and flattened, and protrudes into the spinal canal through the foramen magnum; it may be associated with spina bifida occulta meningomyelocele and other defects. [syn: Arnold-Chiari malformation or syndrome]

arrhinencephaly - Congenital absence of the rhinencephalon and external olfactory organs. (Absence of the olfactory brain and nose). [syn: arhinencephalia]

arthrogryposis - Persistent flexure or contracture of joints.

arthrogryposis multiplex congenita - A generalized development of musculature with contracture and deformity of most joints. See amyoplasia congenital.

arthro-onychodysplasia - See nail patella syndrome.

asphixating thoracic dystrophy - Type of dwarfism associated with respiratory insufficiency and renal failure and other functional renal abnormalities including concentrating ability and impaired proximal reabsorption of bicarbonate, phosphate, amino acids and glucose. [syn: Jeune's syndrome]

astomia - Absence of mouth opening.

asymmetric shaped sternebra - Sternebrae not ossified in a normally symetrical pattern.

asymmetric vertebral body or centrum - Asymmetric incomplete ossification of a vertebral body.

ataxia - Incoordination of muscular action.

atelectasis - Failure of lung to inflate after birth.

atelencephaly - Incomplete development of the brain.

atelo- - Prefix denoting imperfect or incomplete.

atelocardia - Incomplete development of the heart.

atelophalangy - Absent, imperfect or incomplete development of phalanges most distal to metacarpals or tarsals.

atlas - First cervical vertebrae (centrum is usually bipartite in rabbit).

atresia - Imperforation or failure of a normal opening to close.

atreto- - Pertaining to atresia, prefix signifying absence of an opening.

atretocephalus - Imperforate nostrils and mouth.

atretocystia - Bladder atresia.

atretogastria - Stomach atresia.

atretolemia - Larynx, esophagus atresia.

atretometria - Uterus atresia.

atretopsia - Iris atresia.

atretorrhinia - Nose atresia.

atretostomia - Mouth atresia.

atretourethria - Urethra atresia.

***atrial septal defect** - Congenital cardiac abnormalities of the patency of atrial septum due to failure of fusion between either the septum secundum or the primum and endocardial cushions. [syn: the ostium secundum defect]

atrichia - Absence of hair, lacking cilia or flagella.

atricholis congenitalis - Congenital absence of hair.

***atrioventricular canal** - The common canal connecting the primative atrium and ventricle. It normally divides into a right and left orifice, but may persist with other defects.

audiogenic sequor or seizure - Sound-related seizure.

axis - Center line of body. Second cervical vertebrae or epistropheus which bears the odontoid process (dens) about which the atlas rotates.

B

***back knee** - See genu recurvatum.

***balanic or balantic hypospadias** - Commonest type of hypospadias. The urethral opening is at the site of the frenum (the fold on the lower surface of the prepuce or foreskin). The frenum may be rudimentary or absent. A blind pit is in place of the normal urinary opening on the glans penis. [syn: glandular hypospadias or primary hypospadias]

ballooning - Air-filled cavity, such as stomach, intestine; extreme abdominal distention.

barbering - Discrete hairless area.

bent rib - A rib or ribs, which may be bent at various angles and places along the rib axis.

bifurcated - A partial split into two areas of ossification.

bifurcate tongue - Tongue separated into two parts, forked tongue.

bipartite and misaligned sternebrae - The area of ossification is split into two areas of the site. The area between the ossifications is unossified, and the two areas do not line horizonally to the sternum axis. The misaligned ossified area may be fused to another sternebra.

bipartite sternebrae - The area of ossification is split into two areas of the site. There is an unossified area between ossification centers.

bipartite vertebral body or centrum - Ossification of a vertebral body in two separate areas.

birhinia - Double nose.

***blepharelosis** - Turning inward of an edge or margin of the eyelid. [syn: entropion]

***blepharophimosis** - Abnormal narrowness of the palpebral fissures (eye lid opening) in the horizontal direction, caused by displacement of the inner canthi (corner of eye).

***blepharoptosis** - Drooping upper eyelid due to paralysis, ptosis.

***Bochdalek's duct** - See ductus thyroglossus.

***Bochdalek's hernia** - Diaphragmatic hernia where the abdominal contents herniate through the pleuroperitoneal sinus (foramen of Bochdalek).

bone - The formation of bone in development occurs when the ossification has spread across the cartilaginous model.

Bonnevie-Ullrich syndrome - Condition characterized by pterygium colli, lymphangiectatic edema of hands and feet, ocular hypertelorism, short stature, and other developmental anomalies.

***bowleg** - See genu varum.

brachycaudate - Shortened tail. [syn: brachyury]

brachydactyly - Abnormal shortness of digits.

brachygnathia - Shortened lower jaw (see gnathia).

brachyury - Shortened Tail (syn.: brachycaudate).

branched rib - A rib may be branched any where along its axis.

breeding age (female) -

Beagle dog:	9-12 months.
Mouse:	35 to 60 days
Rabbit:	5 to 6 months
Rat:	100 days
Rhesus monkey:	5 yrs.

breeding size (female) -

Beagle dog:	14-16 kg
Mouse:	20 to 30 grams
Rabbit:	4 to 6 kg
Rat:	170 to 200 grams
Rhesus monkey:	10 kg.

***brittle bones** - See osteogenesis imperfecta.

***bronchiectasis** - Chronic dilation of the bronchi marked by fetid breath and paroxysmal coughing with expectoration of mucopurulent matter.

***bulb of heart** - See bulbus cordis.

bulbus arteriosus - See bulbus cardis.

***bulbus cordis** - The foremost of three parts of the primitive heart of the embryo. [syn: bulb of heart and bulbus arteriosus]

buphthalmia - See buphthalmos.

***buphthalmos** - Enlargement and distention of the fibrous coats of the eye. [syn: cornea globosa, megophthalmus, hydrophthalmos, congenital glaucoma and glaucoma of the newborn, infantile glaucoma, macropthalmia, buphthalmus, and buphthalmia]

buphthalmus - See buphthalmos.

C

Caffey's disease or syndrome - Disease of young infants characterized by soft tissue swellings over the affected bones, fever and irritability, and marked by periods of remission and exacerbation. [syn: infantile cortical hyperostosis]

***camptodactyly** - Permanent inward bending of one or more of the fingers.

Camurati-Engelman disease - Characterized by thickening of the cortex of the mid-shaft of the long bones, progressing toward the piphyses, the thickening also sometimes occurring in the flat bones; excessive growth in length of bones of the extremities usually results in abnormal stature. [syn: diaphyseal dysplasia and Englemann's disease]

***canal of Nuck** - Processus vaginalis peritonei. See Nuck's diverticulum.

***canal of Oken** - See Wolffian duct.

***cardia equina** - Collection of spinal roots that descend from the lower part of the spinal cord and occupy the vertebral canal below the cord.

cardiomegaly - Enlargement of the heart (syn.: megalocardia).

***carpus curvus** - See Madelung's deformity.

***cataract** - Opacity of the (crystalline) eye lens.

celoschisis - Fissure of the abdominal wall.

celosomia - Fissure or absence of the sternum with hernial protrusion of fetal viscera.

centra hypoplasia - Incomplete ossification of any one of the vertebral bodies.

cephalocele - Protrusion of brain (cranial contents) through the cranium.

***cephalopagus** - See craniopagus.

cephaly - Pertaining to the head. Cephalo- combining form denoting head.

***cerebroretinal angiomatosis** - See von Hippel-Lindau disease.

cervical arches - There are 7 cervical arches. More than 7 cervical arches are considered to be a variation.

cervical rib - Extra rib arising from cervical vertebra, generally the 7th cervical vertebra.

cervical vertebrae - First seven bones of the spine.

cheilognathopalatoschisis - Cleft in the hard and soft palate, upper jaw, and lip. [syn.: cheilognathouranoschisis]

cheiloschisis - Harelip, unilateral, bilateral, or central [syn: cleft lip]

***chicken chest** - See pectus carinatum. [syn: pigeon breast]

chondrodystrophia calcificans congenita - See Conradi's disease.

chondrodystrophia congenita punctata - See Conradi's disease.

chondrodystrophia fetalis - See achondroplasia.

chondrodystrophia fetalis calcificans - See Conradi's disease.

chondrodystrophy - Morbid condition characterized by abnormal development of cartilage and epiphyses of long bones. General size reduction, limbs disproportionally reduced proximally.

chondroectodermal dysplasia - Achondroplasia occurring in association with defective development of the skin, hair and teeth, polydactyly, and defect of the cardiac septum.

chondro-osteodystrophy - See Morquio's syndrome.

chordee - Band of fibrous tissue associated with hypospadias which causes downward curving of the penis during erection.

*chromosomal analysis -
Group A: Chromosomes 1,2,3.
Group B: Chromosomes 4,5.
Group C: Chromosomes 6-12.
Group E: Chromosomes 13-15.
Group F: Chromosomes 16-18.
Group G: Chromosomes 21,22.

clawfoot - See pes cavus (exaggerated height of the longitudinal arch of the foot.)

cleft lip - See cheiloschisis.

cleft palate - Congenital failure of fusion between right and left palatal processes. Often associated with harelip.

cleido- - Relating to the clavical. Used interchangeably with clido-.

*cleidocranial dysostosis - A rare hereditary condition in which there is defective ossification of the cranial bones, with large fontanels and delayed closing of the suture, complete or partial absence of the clavicles,

and other anomalies. [syn: cleidocranial dysplasia, clidodocranial dysostosis]

***cleidocranial dysplasia** - See cleidocranial dysostosis.

clido- - Relating to the clavical. Used interchangeably with the prefix cleido-.

clinodactyly - Permanent bending or deflection of one or more fingers.

club foot - See talipes.

coarctation - Compression of the walls of a vessel. Narrowing, or contractation as in contractation of an artery.

coclioschisis - Congenital fissure of the abdomen.

collodion baby - See lamellar exfoliation of the newborn.

coloboma - Ocular defect, usually from the failure in a fissure to close. Absence of or defect in a part of the lens, iris, cilary body, or choroid coat. Means the part removed in a mutilation.

coloboma of the choroid - Fissure of the choroid coat from the persistence of a fetal fissure and causing a scotoma on the retina.

coloboma of iris - Fissure of the iris, usually the lower portion.

coloboma lentis - The periphery of the eye lens is incomplete or indented.

coloboma lobuli - Congenital or acquired fissure of the ear lobe.

coloboma of the optic nerve - Defect attributed to a failure of a fetal fissure of the optic stalk to close. Defect attributed to a closure failure of the optic cup.

coloboma palpebrale - A verticle fissure in the eye lid.

coloboma of retina - Congenital fissure in the retina atrributed to incomplete closure of a fissure in the optic cup. [syn: c. of retinae]

***common truncus** - Heart defect; the pulmonary artery and the aorta both arise from a common arterial trunk.

***compression facies** - Facial appearance associated with bilateral agenesis, or hypoplasia of the kidney with increased space between the eyes, a prominent epicanthal fold, flattening of the tip of the nose, prominent crease below the lower lid and low set ears. [syn: Potter's facies]

***congenial ectodermal defect** - See ectodermal dysplasia.

***congenital glaucoma** - See buphthalmos.

***congenital ichthyosiforma erythroderma** - Condition now recognized as two distinct diseases, one bullous (with dominant inheritance) and the other non-bullous (with recessive inheritance). [syn: erythroderma ichthyosiformá (fish scale skin) congenitum]

***conjoined twins** - Duplication of a fetus, ranging from two well-developed individuals joined by superficial connection of varying extent, usually in the frontal, transverse or sagittal body plane (symmetrical conjoined twins) to those in which only a small part of the body is duplicated, or one small and incompletely developed component, the parasite, is attached to a much larger and more fully developed one, the autosite (asymmetrical conjoined twins). Also see craniopagus, dicephalus, pygopagus, thoracopagus and xiphopagus.

Conradi's disease - A rare hereditary condition marked by radiographic appearance of multiple punctate opacities (stippling) in the epiphyses,

usually present at birth and by dwarfism, flexion contractures, cataract, dulled intellect, short blunt fingers, and general weakness. Frequently infants are stillborn or die of associated anomalies in the first year. [syn: chondrodystrophia calcificans congenita, chondrodystrophia calcificans congenita punctata (punctate), chondrodystrophia fetalis calcificans, stippled epiphyses, and dysplasia epiphysealis punctata]

convoluted ureter - Ureter abnormally folded on itself or twisting of the ureter.

coprophagia - Eating of feces. [Syn. rhypophagy, scatophagy]

cor biloculare - Two-chambered heart resulting from the failure to form atrial and ventricular septums.

cor triatriatum - Defect caused by the failure to incorporate the embryonic common pulmonary vein into the left atrium, the pulmonary veins emptying into an accessory chamber superior to the true left atrium and communicating with it by a small opening that obstructs pulmonary venous flow, thus simulating mitral stenosis. [syn: triatrial heart]

cor triloculare - Three-chambered heart.

***cornea globosa** - See buphthalmos.

***coronal hypospadias** - Abnormal opening of the urethra into the corona. The corona is the rim around the proximal part of the glans penis.

corpora albicantia - Plural of corpus albicans.

corpora lutea - (plural) Progesterone secreting yellow bodies found in the ovaries after rupture of Graafian follicles and subsequent expulsion of the ovum. Corpora lutea last for approximately three days after ovulation in unfertilized in rats, and longer in pseudopregnancy, but otherwise throughout gestation and lactation.

corpus albicans - (singular) White fibrous tissue that replaces the regressing corpus luteum in the ovary after parturition in the rat, and during the last half of pregnancy in the human.

corpus luteum - Singular of corpora lutea.

***corrected transposition of the great vessels** - Transposition of the great vessels with inversion of the ventricles and atrioventricular valves. [syn: mixed levocardia]

***coxa adducta** - See coxa vara.

***coxa flexa** - See coxa vara.

***coxa valga** - See coxa vara.

***coxa vara** - Deformity of the hip (coxa) in which the angle formed by the axis of the head and the neck of the femur and the axis of its shaft is materially decreased. [syn: coxa adducta, coxa flexa and coxa valga]

craniocele - Herniation of the brain through skull. See encephalocele.

craniofacial dysostosis - A hereditary disorder (in humans) characterized by acrocephaly, exophthalmos (abnormal protrusion of the eyeball), hypertelorism (abnormal distance between "eyes"), strabismus (disorder of the eye in which optic axes cannot be directed to the same object), parrot-beaked nose, and hypoplastic maxilla with relative mandibular prognathism (projection of the jaws beyond projection of forehead). [syn: Crouzon's disease]

***craniopagus** - Conjoined twins united by the heads. [syn: cephalopagus]

craniorachischisis - Fissure of cranium and spinal cord. Rachischisis (spina bifida) which extends into the cranium. It may occur with anencephaly.

craniorrhachischisis - Congenitally unclosed skull and spinal column.

***craniorrhachischisis totalis** - Exposure of the entire central nervous system.

cranioschisis - Congenital fissure of the cranium.

***craniosynostosis** - Premature closure of one or more sutures of the skull.

cranium bifidum - Congenital cleft of the cranium.

crazy chick disease - See encephalomalacia.

cri-du-chat syndrome - A hereditary congenital syndrome characterized by hypertelorism, microcephaly, severe mental deficiency, and a plaintive cat-like cry. Caused by deletion of the short arm of a chromosome in Group B (chromosome 4 or 5).

Crouzon's disease or syndrome - See craniofacial dysostosis. [syn: hypertelorism]

cryptorchidism - Developmental defect whereby the testes are located anterior to the bladder instead of lateral (failing to descend into the scrotum). May be unilateral or bilateral. [syn: cryptorchism]

***cutis elastica** - See Ehers-Danlos syndrome.

***cutis hyperelastica** - Hyperelastic skin. See Ehers-Danlos syndrome.

cyclopia - Single eye or closely approximated eyes with all intergrades in a single orbit

cyst of fimbriae - the fimbria is a tube-like or funnel-like structure with proximal end at the ovary and distal end joining with the fallopian tube, which opens into the uterine cavity. [syn: the infundibulum of the uterine tube, and the fimbriae cyst]

D

dactyly - Digit.

dactylomegaly - Abnormal large digits.

Danlos's syndrome - See Ehlers-Danlos syndrome.

decidua - Endometrial lining of the uterus thickened and altered for the reception of the fertilized ovum. It is shed when pregnancy terminates.

delayed ossification - Incomplete ossification of an otherwise normal center.

developmental toxicity - Adverse effects on the developing organism which may result from exposure prior to conception (either/both parents), during prenatal development (as in the FIFRA Guideline Study, §83-3) or postnatally to the time of sexual maturation. However, adverse developmental effects may be detected at any point in the lifespan of the organism. The end points of developmental toxicity include: 1) death of the developing organism, 2) structural abnormality (malformations and variations), 3) altered growth, and 4) functional deficiency (See Section III. B., below).

dexiocardia - See dextrocardia

dextrocardia - Transposition of the heart to the right side of thorax, with the apex pointing to the right, occurring with transposition (situs inversus) of the abdominal viscera, or without such transposition (isolated dextrocardia). [syn.: dexiocardia].

diaphragmatic eventration - Elevation of the dome of the diaphragm, usually the result of paralysis of the phrenic nerve.

diaphragmatic hernia - Protrusion of abnormal viscera into the thoracic cage through a defect in the diaphragm [syn.: diaphragmatocele].

diaphragmatocele - See diaphragmatic hernia.

***diaphyseal dysplasia** - See Camurati-Engelman disease.

***diastematomyelia** - A congenital defect often associated with spina bifida, in which the spinal cord is split by a bony spicule or a fibrous band, each half being surrounded by a dural sac.

***diastrophic** - Dwarfism associated with talipes equinovarus (clubfoot), scoliosis, and "hitchhikers" deformity of the hand. Inherited as an autosomal recessive trait.

***dicephalus** - Conjoined twins with two heads and one body. There are various degrees of separation. Dicephalus dipus dibrachius has two heads, two feet, and two arms. Dicephalus dipus tetrabrachius has two heads with varying degrees of fusion of the upper trunk, each component having two heads, two feet and four arms. Dicephalus tripus tribachius has two heads, three feet, and three arms. There are other combinations.

dilatation of the renal pelvis - Dilatation of cranial end of the ureter into which urine is excreted from neonatal kidneys. It is manifested by an increase in the relative size of the pelvis compared with the papilla. May be accompanied by dilated ureters, and obstructed urine flow. A variation that may disappear as the neonate matures, or develop into decreased renal function.

diplomyelia - Lengthwise fissure and seeming duplication of the spinal cord.

disk or donut kidney - Fusion of both upper and lower poles.

displaced, rudimentary - A small ossification site that may be displaced from the normal position.

displaced vertebral body or centrum - Ossification of vertebral body is displaced from the line through the arches.

distended bladder - Expanded or swollen bladder that may be the result of an obstruction.

distended ureter - Ureter enlarged or swollen, possibly the result of fluid accumulation caused by a blockage. [syn: hydroureter]

***dolichocephalic, dolichocephalous** - Long headed; having a cephalic index of 75.9 or less. [syn: mesocephalic]

***dolichocephaly** - The quality of being dolichocephalic.

dolichodactyly - Abnormal long digits.

***dolichostenomelia** - See arachnodactyly.

domed head - Dome-shaped head, with or without hydrocephaly.

double ureters - Characterized by a doubling of a portion of one ureter, due to incomplete fusion.

Down's syndrome - Characterized by a small, anteroposteriorly flattened skull, short phalanges, with moderate to severe mental retardation. [syn: mongolism and trisomy 21 syndrome]

***duct of His or Vater** - See ductus thyroglossus.

***duct of Wolff** - See Wolffian duct.

ductus arteriosus - A fetal blood vessel connecting the pulmonary artery directly to the descending aorta in the fetus; atrophies after birth.

ductus mesonephricus - See Wolffian duct.

ductus thyroglossus - A duct in the embryo extending between the thyroid primordium and the posterior part of the tongue. [syn: duct of His or Vader, Bochdalek's duct, His's Canal, and thyrolingual duct]

ductus wolffi - See Wolffian duct.

dumbell asymmetric - The two ends of the dumbell shaped ossification area are not symmetrical.

dumbell shaped sternebra - Sternebra ossified in the shape of a dumbell.

dumbell vertebral body or centrum - Ossification of the vertebral body in the form of a dumbbell.

***dysautonomia** - A hereditary condition characterized by defective lacrimation, and hyporeflexia (weakened reflexes). [syn: familial autonomic dysfunction]

dysmelia - Malformation of a limb including excessive development as well as reduction deformities.

***dysostosis enchondralis epiphysaria** - See dysplasia epiphysealis multiplex.

dysplasia - Abnormal tissue development.

***dysplasia epiphysealis multiplex** - A developmental abnormality of various epiphyses, which appear late and are mottled, flattened, fragmented, and usually hypoplastic. [syn: dysostosis enchondralis epiphysaria and multiple epiphysealis dysplasia]

***dysplasia epiphysealis punctata** - See Conradi's disease.

E

***eccentro-osteochondrodysplasia** - See Morquio's syndrome.

ecchymosis - Purple patch caused by extravasation of blood into the skin or a small hemorrhagic spot (syn.: hematoma).

ectocardia - Exposure of the heart due to insufficient development of the wall of the chest. Displacement of the heart outside of thorax.

***ectodermal defect congenita** - See ectodermal dysplasia.

***ectodermal dysplasia** - A rare hereditary condition affecting chiefly males and marked by a smooth glossy skin, total absence of sweat glands, abnormality of the teeth, and defective hair formation. [syn: ectodermal defect congenita]

ectopia - Displacement or malposition.

ectopia cordis - See ectocardia.

***ectopia vesicae** - See exstrophy of the bladder.

ectopic - Located away from the normal position.

ectopic kidney - Abnormal position (e.g., pelvic kidney).

ectopic ureters - An abnormally placed ureter opening, either into the urinary bladder or at another site lower in the urinary or genital tract.

ectrodactyly - Fewer digits than normal [syn: perodactyly, oligodactyly]

ectromelia - Absence of a limb or limbs (see melia).

*ectropion - The turning outward (eversion) of an edge or margin, as of the eyelid, resulting in exposure of the palpebral conjunctiva.

ectrosyndactyly - Absence of some digits with remaining digits fused or webbed.

Edward's syndrome - Condition characterized by neonatal hepatitis, mental retardation, scaphocephaly or other skull abnormality, micrognathia, blepharoptosis, low-set ears, corneal opacities, deafness, webbed neck, short digits, ventricular septal defects, Meckel's diverticulum, and other deformities. [syn: trisomy E syndrome and trisomy 18 syndrome]

Ehlers-Danlos syndrome - Characterized by hyperextensibility of the joints and hyperelastisity and fragility of the skin with poor healing of wounds, leaving scars resembling parchment, by capillary fragility, and by subcutaneous mucinous or fatty nodules following trauma. [syn: Danlos's syndrome, cutis elastica, cutis hyperelastica, elastic skin, and India rubber skin]

Eisenmenger's syndrome - Ventricular septal defect with pulmonary hypertension and cyanosis due to right-to-left (reversed) shunt of blood.

elastic skin - See Ehler's-Danlos syndrome.

embryo - The developing organism from the fertilized ovum through major organogenesis. The approximate time of completion for major organogenesis is as follows for selected species (days after fertilization):

Hamster:	day 14
Mouse:	day 16
Rabbit:	day 18
Rat:	day 16

embryotoxicity - Toxic effects on the conceptus which include any of the four indicators of developmental toxicity; 1) death, 2) structural abnormality, 3) altered growth and, 4) functional deficit. Once used to describe early resorption, and structural abnormality, but now considered to be developmental toxicity because of the problems distingushing between embryo and fetal toxicity.

encephalocele - Protrusion of brain substance through the skull but the cerebrum is well formed and is covered by connective tissue; skin-covered brain.

encephalomalacia - Softening of the brain, with functional deficit in chickens. [syn: crazy chick disease]

encephaly - Pertaining to the brain.

*endocardial cushion defect - A spectrum of septal defects resulting from imperfect fusion of the endocardial cushions in the embryonic heart, and ranging from persistent ostium primum to persistent common atrioventricular canal. In the most severe form there is a large defect in the lower part of the atrial septum, a defect in the upper part of the ventricular septum, and both the tricuspid and mitral valves are deformed.

*endocardial cushions - Elevations on the atrioventricular canal of the embryonic heart. In abnormal development they fuse with the free edge of the septum primum to separate the right and the left atria.

endotheliochorial placenta - A placenta in which the syncytial trophoblast embeds maternal vessels bared to the endothelial lining, which is in contact with the chorion of the fetal membranes. Species with this placenta type are the dog, cat, ferret, bear, most bats, and the tree shrew.

Englemann's disease - See Campurati-Engelman disease. [syn: diaphyseal dysplasia]

***entropion** - The turning inward (inversion) of the edge or margin, as of the margin of the eyelid, with the tarsal cartilage turned inward toward the eyeball. [syn: blepharelosis]11

***epidermolysis bullosa** - A group of genetically transmitted diseases of the skin, marked by the development of bullae and vesicles, often at the site of trauma; the group includes e. bullosa dystrophica, e. bullosa simplex, and probably other variants. [syn: epidermolysis bullosa hereditaria]

***epidermolysis bullosa hereditaria** - See e. bullosa.

***epispadias** - A congenital defect of the penis. The urethra opens on the dorsum of the penis.

epistropheus - Second cervical vertebrae [syn.: axis].

epitheliochorial placenta - A type of placentation in which the uterine lining is not eroded but merely lies in apposition. The chorion is in contact with the uterine epithelium. Species with type of placenta are the cow, ewe, goat, horse, pig, tapiar, American mole, dolphin, and whale.

***erythrodermaichythyosiforme congenitum** - See congenital ichythyosiforme erythroderma.

esophageal stenosis - Constriction of esophageal lumen.

estrous cycle - Cycles of sexual receptivity in female mammals (other than primates). The cycles are divided on the basis of ovarian activity into estrus (period of sexual activity and proximity to follical rupture or ovulation), metestrous (early development of the corpus luteum), diestrus (mature corpus luteum), and proestrus (period of follicular activity). [syn: oestrous cycles] Also refer to ovulation time.

Rats (4-5 day cycle)

Proestrus	≈12 hours, round-oval cells.
Estrus	≈12 hours, maximal fluid distension, cornified cells.
Metestrus	≈21 hours, cornified cells & leucocytes.
Diestrus	≈57-70 hours = 2.4-2.9 days, leucocytes only.

Beagle dogs (5-12 month cycle)

Proestrus	≈4-14 days.
Estrus	≈4-14 days.
Metestrum	≈ 60 days.
Anestrum	90+ days.

estrous cycle recurrence -

Beagle Dog:	≈5-12 months
Guinea Pig:	≈13-18 days
Hamster:	≈4 days
Mouse:	≈4-5 days
Rabbit:	polyestrus
Rat:	≈4-5 days

estrus - Period in the estrous cycle of female mating receptivity. [syn: oestrus]

exencephaly - Congenital exposure of the brain due to absence of cranium. Disorganized outward growth of brain.

***exfoliation** - Falling off of scales or layers.

exocardia - Abnormal position of the heart.

exomphalos - Congenital condition due to failure of development of the abdominal wall. The intestines protrude through a gap in the umbilical region, covered only by a thin membrane [syn.: umbilical hernia].

exophthalmos - Protrusion of eyeball [syn.: exphthalmia]. Also see ophthalmia.

***exostosis** - A benign bony growth projecting outward from the surface of a bone, characteristically capped by cartilage.

external hydrocephaly - Fluid accumulated in subarachnoid space over the brain and under the dura matter.

extra kidney - More than two kidneys, the extra kidney must be associated with an extra ureter.

extra cervical vertebrae - The presence of a extra cervical vertebrae, generally called the 8th cervical vertebrae.

extra ossification center - Extra site of ossification between two normal sites, which may or may not be ossified.

extra ribs - 14th ribs, greater than half the length of 13th thoracic ribs.

extra thoracic body or centrum - A 14th thoracic centrum can occur in addition to the 13 normally present.

***extrophy of the bladder** - A deficiency of the abdominal wall and bladder; the latter organ appears to be turned inside out, having the internal surface of the posterior wall showing through the opening in the anterior wall. [syn: ectopia vesicae.]

F

***familial autonomic dysfunction** - See dysautonomia.

***familial ostestchondrodystrophy** - See Morquio's syndrome.

fertilization - Impregnation of an ovum by a spermatozoon.

fetal wastage - Post implantation loss.

fetotoxicity - Toxicity in fetuses detected between the end of major organogenesis and parturition. It may be any of the four indicators of developmental toxicity; 1) death, 2) structural abnormality, 3) altered growth and , 4) functional deficit. Once used to describe fetal death, weight decrement, size decrement, delayed ossification, and effects on the developing testes and brain, but now considered to be developmental toxicity because of the problems distinguishing between toxicity to embryo and fetus without special studies.

fetus - The developing organism from the period of major oranogenesis to parturition.

fifteenth - Lumbar rib, considered as malformation.

fimbria - Any fringe like structure.

fimbrial cyst - Cyst of fimbriae, the fimbria is a tube-like or funnel-like structure with proximal end at the ovary and distal end joining with the fallopian tube, which opens into the uterine cavity. [syn: infundibulum of the uterine tube]

***flatfoot** - See pes planus and pes valgus.

fontanelle - Space at the junction of the occipital and two parietal bones. Ossifies after birth.

food consumption (average) - Rule of thumb estimations, which will vary with animals weight and age.

Rat, weanling: 9 grams/day
Rat, adult: 15 grams/day
Mouse, weanling: 5 grams/day
Mouse, adult: 7 grams/day

***foramen ovale** - An opening between the left and right atria of the heart, which normally closes after birth. [syn: patent ovale foramen]

foramen ovale cordis - See foramen ovale.

fourteenth rib - 14th rib or ribs present (13 ribs normally present in the rat). The difference between a 14th rib and 14th rudimentary rib is generally defined by the investigator. [Syn: supernumerary rib]

fused arches - One or more vertebral arches fused together.

fused arches and body or centrum - Vertebral arches are fused through the vertebral centrum to each other at the same vertical position.

fused body or centrum - Centra fused together.

fused ribs - Two or more ribs, which may be fused distally, medially or proximaly along the axes of the ribs.

fused sternebrae - Two sternebrae fused together.

***fragilitas ossium** - See osteogenesis imperfecta.

Franceschettis's syndrome - See Treacher-Collins syndrome.

Francois' syndrome - See Hallerman-Streiff syndrome.

functional deficit - A general term used to mean any adverse effect on the ablilty of the organism to function including adverse effects on physiology or behavior.

functional developmental toxicology - Study of the causes, mechanisms, and results of alterations in functional competence of the organism after exposure during critical periods of development.

***funnel breast or chest** - See pectus excavatum.

fused kidney - Union of the extremities (usually lower) of the two kidneys by a band of tissue extending across the vertebral column.

fusion - The merging or coherence of adjacent parts or bodies.

G

Gardner's syndrome - Familial polyposis of the large bowel (with malignant potential), supernumerary teeth, fibrous dysplasia of the skull, osteomas, fibromas, and sebaceous cysts.

gastromegaly - Enlargement of the stomach.

gastroschisis - Congenital fissure in the abdominal wall not including the insertion cite of the umbilicus, and usually accompanied by protrusion of the small and part of the large intestine. Congenital opening of the abdomen.

***genu recurvatum** - Hyperextension of the knee. [syn: back knee]

***genu valgum** - A deformity in which the knees are abnormally close together and the space between the ankles is increased. [syn: knock-knee]

***genu varum** - A deformity in which the knees are abnormally separated and the lower extremities are bowed inwardly; the deformity may be in the thigh or the leg, or both. [syn: bowleg]

***gerbode defect** - A type of ventricular septal defect where there is a fault in the part of the septum between the ventricle and the right atrium frequently with tricuspid valve involvement.

gestation - The period of development in viviparous animals from the time of fertilization of the ovum to parturition.

gestation day 16 - Considered in rats and mice as the time separating embryonic and fetal period of development.

***glandular hypospadias** - See balanic or balanic hypospadias.

glaucoma of the newborn - See buphthalmos.

glossa - Tongue.

***glossoptosis** - Downward displacement or retraction of the tongue.

gnathia - Jaw, mandible.

Goldenhar's disease or syndrome - Congenital condition in which colobomas of the upper eyelid, epibulbar dermoids, bilateral accessory auricular appendages anterior to the ears, and vertebral anomalies are frequently associated with characteristic facies, consisting of asymmetry of the skull, prominent frontal bossing, low hairline, mandibular hypoplasia, low-set ears, and sometimes, hemifacial microstomia. [syn: oculoauricular dysplasia, mandibulfacial dystosis with epibulbar dermoids, OAV syndrome, and oculoauriculovertebral (OAV) dysplasia]

***gonadal dysgenesis** - A general term for a variety of gonadal development anomalies, including gonadal aplasia, Turner's syndrome, hermaphroditism, and psuedohermaphroditism.

Graft-verses-host disease - A disease chactacterized by an in-utero infusion of maternal blood and immune cells into the fetus, causing graft-verses-host-disease in the offspring or a runt, weak offspring of low birth weight or offspring with poor survival. Whether or not this mechanism applies to all runting has not been established. Also see runt and weight, Maximum Stunted (MSW).

gravid - Pregnant, containing developing young.

gravida - Description used to indicate the number of pregnancies as in gravida I or primigravida, or gravida II, etc. for humans.

great vessels anomalies - Any developmental defect of the great vessels (for example: two left carotid arteries, retro-tracheal and/or retroesophageal aorta, missing right subclavian artery, reversed aorta, pulmonary artery stenosis, left common carotid artery arising from the innominate artery, etc.).

growth Index - Average weights of male and female offspring at birth, 4, 7, 14, or 21 days of age.

Gunn's syndrome - See jaw-winking syndrome.

H

Hallermann-Streiff syndrome - Characterized by dyscephaly (usually brachycephaly), parrot nose, mandibular hypoplasia, proportionate nanism, hypotrichosis, bilateral congenital cataracts, and microphthalmia. [syn: Hallermann-Francois syndrome, Francois' syndrome, mandibulo-oculofacial dyscephaly and oculomandibulofacial syndrome]

***hallux valgus** - Angulation of the great toe toward the other toes; the great toe may ride under or over the other toes.

***hallux varus** - Angulation of the great toe away from the other toes.

***harlequin fetus** - A fetus covered with thick horny, armor-like plates. The severest form of lamellar exfoliation of the newborn (see lamellar exfoliation); it is usually stillborn or dies shortly after birth.

hematoma - [syn.: ecchymosis (small irregular hemorrhagic area, but larger than a petechia of the skin)].

*hemianencephaly - Congenital absence of one side of the brain.

hemimelia - Absence of all or part of the distal portion of a limb (see melia).

hemiplasic - Partially formed.

hemisternebrae - See unilateral sternebral ossification.

hemivertebra - Defect of one half of a vertebra.

hemochorial placenta - A type of placenta in which maternal blood comes in direct contact with the chorion. Species with this type of placenta are some rodents, man, rhesus, ape, baboon, armadillo, three tailed and mastiff bat, and some shrews.

hemoendothelial - A type of placenta so attenuated that maternal blood appears to be separated from fetal blood only by the endothelium of the chorionic capillaries. Species with this type of placenta is the rabbit. Also see placenta.

hermaphrodite - Offspring with both male and female gonads and characteristics. Also see psuedohermaphrodite.

hermaphroditism - See gonadal dysgenesis.

hernia - Protrusion of a loop or knuckle of an organ or tissue through an abnormal opening.

heterotopia - Normal tissue in an abnormal location.

Hippel's disease - See von Hippel-Lindau disease.

*His' canal - See ductus thyroglossus.

histogenesis - Period of prenatal development that slightly overlaps organogenesis but extends primarily into the fetal period.

*holoprosencephaly - Failure of cleavage of part of the brain (prosencephalon) with absence of other portions of the brain. Also a deficit occurs in the midline facial development that can result in cyclopia, and a large proboscis with cleft lip and palate. It is associated with trisomy 13, 15, or 18 and other cytogenic anomalies. See Patau's syndrome.

*honeycomb lung - The appearance of multiple small radiolucent shadows on the lung X-ray, representing dilations of smaller, less rigid airways or multiple small cysts or cavities.

horseshoe kidney - Fusion of lower poles (common) or fusion of upper poles (rare).

hump back - See kyphosis.

hunch back - See kyphosis.

*hydatid of Morgagni - A cyst-like remnant of the Mullerian duct attached to a testis or to the oviduct. [syn: morgagnian cyst]

hydramnios - Excess amniotic fluid (maternal defect).

*hydranencephaly - Complete or almost complete absence of the cerebral hemispheres, the space they normally occupy being filled with cerebrospinal fluid. [syn: internal hydrocephalus]

hydrocele - A swelling due to accumulation of serous fluid, especially a collection of fluid in the tunica vaginalis of the testicle or along the sperm cord.

hydrocele on oviduct - Cystic ovaries.

hydrocephaly - Marked enlargement of the ventricles of the cerebrum due to excess cranial cerebrospinal fluid (CSF). Inadequate drainage of CSF during embryonic development. Classified as slight (sometimes referred to as

dilated brain ventricles), moderate (enlargementof ventricles with little or no thinning of the brain), or severe (enlargement of ventricles with thinning surrounding brain regions).

hydrometra - Watery fluid within the uterus. Normally occurring at proestrus and maximally at estrus.

***hydromyelia** - A pathological condition characterized by abnormal accumulation of fluid in the enlarged central canal of the spinal cord or elsewhere in the cord substance.

hydronephrosis - Abnormal distention of renal pelvis with accumulation of fluid due to obstructed outflow (may be accompanied by hydroureter). Obstruction can be due to epithelial hyperplasia in the ureteric lumen (true hydronephrosis) or to delayed ureteric membrane degeneration (apparent hydronephrosis) See renal papillae and renal pelvis.

***hydrophthalmos** - See buphthalmos.

hydrosalpinx - Distention of the fallopian tube with watery fluid

hydrothorax - Presence of fluid in the pleural cavity.

hydroureter - Distention of ureter with urine or fluid. [syn: distended ureter]

hyoid body - Small ossification center at the ventral midline of the neck (observed in neonates; it is the presumptive hyoid bone in adults).

hyoid bone - U-shaped bone at the root of the tongue

***hypermobile flatfoot** - See pes valgus.

hyperplasia of the kidney - Enlarged kidney due to abnormal multiplication of cells in the kidney.

hypertelorism - Abnormal distance between two paired organs (e.g., ocular hypertelorism).

***hypoplasia** - Incomplete development of an organ so that it fails to reach adult size; it is less severe in degree than aplasia.

***hypoplasia of the mesenchyme** - See osteogenesis imperfecta.

***hypospadias** - The urethra opens on the underside of the penis or on the perineum. See also balanic h., coronal h., penile h., penoscrotal h., and perineal h.

I

***ichthyosis** - Any of several skin disorders characterized by dryness, roughness, and scaliness due to hypertrophy of the horny layer, resulting from excessive production or retention of keratin, or a molecular defect in keratin. Also see lamellar exfoliation in the newborn.

***ichthyosis, bullous type** - A scaling skin disease characterized by blistering with autosomal dominant inheritance. The condition is apparent in the first 6 months of life.

***ichthyosis congenita** - See lamellar exfoliation of the newborn.

***ichthyosis fetalis** - A hereditary form of ichthyosis transmitted as an autosomal dominant or sex-linked recessive trait. [syn: ichthyosis simplex] See also lamellar exfoliation of the newborn.

***ichthyosis X-linked** - A scaling disease affecting males only, that is apparent in the first year of life.

***india rubber skin** - See Ehlers-Danlos syndrome.

***infantile cortical hyperostosis** - See syndrome: Caffey's.

infantile glaucoma - See bluphthalmos.

infundibulum of the uterine tube - A tube-like or funnel-like structure with proximal end at the ovary and distal end joining with the fallopian tube, which opens into the uterine cavity. [syn: cyst of fimbriae]

***iniencephaly** - A developmental anomaly characterized by enlargement of the foremen magnum and absence of the laminal and spinal processes of the cervical, dorsal, and sometimes lumbar vertebrae, with vertebrae reduced in number and irregularly fused, the brain and much of the spinal cord occupying a single cavity.

innominate artery absent - Condition wherein the right subclavian and common carotid artery originate directly off the aortic arch instead of branching off a common innominate artery

insemination, artificial (rabbit) - Procedure:

a) Collecting Apparatus: artificial vagina filled with warm water (55 degrees C) and fitted with a graduated collecting tube.

b) Removing of Semen: after collection, the gelatinous plug is removed and the ejaculate evaluated for volume, sperm motility, and concentration.

c) Volume: determined from the graduated collecting tube.

d) Motility: determined microscopically on a percentage basis from a slide using low power magnification.

e) Concentration: determined using a standard dilution in a red blood cell pipette and a hemacytometer and examined under higher power magnification.

f) Insemination: deposits 0.2 to 0.5 ml/aliquot of diluted semen (25% in normal saline) into the anterior vagina with a glass insemination pipette.

g) hCG injection: human chorionic gonadotropin (100 USP) injection via the marginal ear vein immediately following insemination to ensure ovulation.

h) Precaution: diluted semen from each buck should be used to inseminate an equal number of does in each group.

interauricular septal defect - Incomplete closure of the atrial septum.

internal hydrocephaly - Fluid accumulated within the brain ventricles. Term also applied to hydranencephaly when brain is completely or partially undeveloped.

interrupted rib ossification - Intermittent ossification in an otherwise normal rib.

intrauterine deaths and resorptions - Usually classified as early and late. Late deaths show embryonic or fetal tissue in addition to placental tissue, whereas early deaths show decidual or placental tissue only.

irregular shaped sternebra - Xiphisternum may have an irregular shape.

isodactyly - Digits of equal length.

***ivory bones** - See osteopetrosis.

J

***Jansen's disease** - See metaphyseal dysostosis.

jaw-winking syndrome - Unilateral ptosis of the eyelid with movement of the upper eyelid with those of the jaw. [syn: Gunn's syndrome]

Jeune's syndrome - See asphyxiating thoracic dystrophy.

K

Kartagener's syndrome - Hereditary disorder involving a combination of dextrocardia (situs inversus), bronchiectasis and sinusitis, transmitted as an autosomal recessive trait.

keratocentesis - Aqueous paracentesis (surgical puncture of a cavity for aspiration of fluid).. Also see keratonyxis.

keratonyxis - Puncture the cornea; specifically, the operative puncture with a view to couching or needling the lens in cataract. Also see keratocentesis.

Klinefelter's syndrome - Condition characterized by the presence of small testes with fibrosis and hyalinization of the seminiferous tubules, impairment of function and clumping of Leydig cells, and increase in urinary gonadotropins; these are males with genotype XXY or XXXY.

Klippel-Feil syndrome - Shortness of the neck resulting from reduction in the number of cervical vertebrae or fusion of multiple hemivertebrae into one osseous mass.

knock knee - See genu valgum.

kyphoscoliosis - Backward and lateral curvature of the spinal column (cord). Sometimes congestive heart failure is a complication. Present in over 1% of cor pulmonale cases.

kyphosis - Excessive convexity in the (backward) curvature of the thoracic (dorsal) spine. [syn: lordosis, scoliosis, hunchback, and humpback]

L

***lamellar desquamation of the newborn** - See lamellar exfoliation of the newborn.

***lamellar exfoliation of the newborn** - A rare congenital disorder transmitted as an autosomal recessive trait, in which the infant (collodion baby) is born completely covered with "collodion", a parchment-like membrane that peels off within 24 hours, after which there may be complete healing, or the scales may reform and the process repeated. [syn: ichthyosis fetalis, lamellar desquamation of the newborn, ichthyosis congenita, and lamellar ichthyosis of the newborn]

***lamellar ichthyosis of the newborn** - See lamellar exfoliation of the newborn.

Larsen's syndrome - Cleft palate, flattened facies, multiple congenital dislocations and foot deformities.

lethargy - Excessive drowsiness.

Leydig's cells - Testicular interstitial cells of Leydig (cells among the seminiferous tubules), which synthesize testosterone in response to luteinizing hormone (LH) stimulation. See also Wolffian duct.

***Leydig's duct** - See Wolffian duct.

***Lindau-von Hippel disease** - See von Hippel-Lindau disease.

***lingua frenata** - See tongue-tie.

***lissencephaly** - See agyria.

litter size - Ratio of total number of viable fetuses/total number of females with viable fetuses.

*lordosis - The anterior concavity in the curvature of the lumbar and cervical spine as viewed from the side.

lumbar arches and centra - There are 6 lumbar arches and centra.

lumbosacral shift - A condition in which the two focal ossification areas on either side of the first sacral vertebrae will be staggered with one ossification adjacent to first sacral vertebrae and the other adjacent to the second sacral vertebrae. The lumbosacral shift is often accompanied by unaligned pelvic bones.

lump kidney - Fusion of both kidneys to form only one kidney.

lung agenesis - Complete absence.

lung agenesis of a lobe - Absence of a lobe (usually the intermediate lobe in rabbits).

lung aplasia - Rudimentary bronchi; absence of pulmonary and vascular structures.

lung, unilobular - Condition in which the lung consists of one lobe instead of several (right lung of the rat has four lobes).

Lutembacher's disease or syndrome - Atrial septal defect (usually of the foramen ovale type) with mitral stenosis (usually rheumatic).

M

*macrocheilia - Excessive size of lips.

macroglossia - Large tongue (see Glossa).

macrognathia - Large jaw (see Gnathia).

macromelia - Abnormally large limbs.

macrophthalmia - Abnormally large eyes.

macrorrhinia - Nose hypertrophy.

macrosomia - Greatly increased body size.

*macrostomia - Greatly exaggerated width of mouth, resulting from failure of the union of the maxillary and mandibular process, with the unilateral or bilateral extension of the oral orifice to the ear.

*macrotia - Abnormal enlargement of the pinna of the ear.

*Madelung's deformity - The radial deviation of the hand secondary to overgrowth of the distal ulna or shortening of the radius. [syn: carpus curvus]

Malaligned sternebrae - See misaligned sternebrae.

malformation - A permanent structural change in an organism, which may adversely affect survival, development, or function.

*mandibulofacial dysostosis - See Treacher-Collins syndrome.

*mandibulofacial dysostosis with epibulbar dermoids - See Golderhar's syndrome.

*mandibulofacial-oculofacial dyscephaly - See Golderhar's syndrome.

manubrium - First sternebrae [syn.: presternum].

*marble bones - See osteopetrosis.

Marfan's syndrome - Abnormal length of the extremities, especially of fingers and toes, subluxation of the lens, cardiovascular abnormalities (commonly dilation of the ascending aorta) and other abnormalities. It is an autosomal dominant trait with variable expression.

maternal wastage - In developmental toxicity studies, usually refers to the sum of aborted, dead, and premature delivery dams.

maximum stunted weight - (See weight).

***medullary cystic disease, juvenile type** - A bilateral cystic disease of the kidneys with multiple cysts in the medulla. This disease results in diabetes insipidus and progressive renal failure.

megalocardia - Enlarged herat. [syn: cardiomegaly]

megaly - Abnormal enlargement (e.g., hepatomegaly).

***megophthalmos** - See buphthalmos or hydrophthalmos.

melia - Limb.

meningocele - Hernia protrusion of membrane enveloping the CNS; skin is intact and translucent and is elevated by a fluid filled vesicle of meninges which protrudes through a midline defect in the cranium or vertebral column.

meningoencephalocele - Hernia protrusion of brain and its enveloping membrane; intact skin but the brain or some part of it together with meninges protrudes through a cranial defect to cause an irregular mass beneath the skin.

meningomyelocele - See myelomeningocele.

mesocephalic - Long headed; having a cephalic index of 75.9 or less. [syn: dolichocephalic, and dolichocephalous]

***metaphyseal chondrodysplasia** - See metaphyseal dysostosis.

***metaphyseal dysostosis** - A skeletal abnormality in which the epiphyses are normal, or nearly so, and the metaphyseal tissues are replaced by masses of cartilage, producing interference with enchondral bone formation, and expansion and thinning of the metaphyseal cortices. [syn: Jansen's disease and metaphyseal chondrodysplasia]

***metatarsus varus** - A congenital deformity of the foot in which its inner border is off the ground with the sole turned inward, the patient walking on the outer border of the foot.

***metatrophic** - Utilizing lifeless organic matter for food. See also paratrophic.

metrial gland - A small pinpoint round hemorrhagic (petechial in appearance) like area of the uterus indicating an implantation site for a delivered offspring, or a site of a resorbed embryo/fetus. They form singly (one resorption) or longitudinally at multiple sites along (multiple resorptions or delivered offspring) the uterine lining. They can be seen if resorption occurs one or two days after implantation. These sites are still visible at necropsy through parturition and weaning. They can be frequently seen even after a second pregnancy where they can confuse the second pregnancy implantation site counts. (syn.: metrial site, metrial scar, implantation site, or implantation scar).

microcephaly - Small head/brain.

***microcheilia** - Abnormal smallness of the lips.

microchiria - Abnormally small hands.

microglossia - Small tongue.

micrognathia - Small jaw (see gnathia).

***microgyria** - A malformation of the brain characterized by development of numerous small convolutions (microgyria). [syn: polymicrogyria]

micromelia - Abnormally small or short limb.

microphthalmia - Abnormal smallness of the eyes.

microstomia - Abnormal smallness of the mouth.

***microtia** - Gross hypoplasia or aplasia of the pinna of the ear with a blind or absent external auditory meatus.

misaligned sternebrae - Two areas of ossification of a sternebra do not line up with axis of the sternum. [syn: malaligned sternebrae]

missing rib - Absence of a rib from part of the vertebral column.

missing sternebrae - The cartilaginous model and ossification are absent from the normal site.

missing vertebral body - The cartilaginous skeleton and ossification at a designated position are absent.

missing vertebral lumbar centra - The absence of one or more lumbar centra.

missing vertebral thoracic arch - A unilateral or bilateral absence of an arch.

***mixed levocardia** - See corrected transposition of the great vessels.

***mongolism** - See Down's syndrome.

monocardian - Heart with only one atrium and one ventricle, as that of a shark.

monorchidism - Only one descended testicle.

monozygotic twins - one fertilized egg divided, two fetuses, one placenta.

***monster** - A fetus or infant with such pronounced developmental anomalies as to be grotesque and usually non-viable. The Greek word "teratos" means monster and is the root of the term teratology.

***morgagnian cyst** - See hydatid of Morgagni. Also a cyst resembling or an echinococcus cyst (a small tapeworm).

Morquio's syndrome - Mucopolysaccharidosis becoming evident when the affected infant walks. [syn: mucopolysaccharidosis IV, eccentro-osteochondrodysplasia, chondro-osteodystrophy, familia, and osteochondrodystrophia deformans]

***mucopolysaccharidosis IV** - See Morquio's syndrome.

***multicystic renal dysplasia** - A cystic renal disease, that may be unilateral or bilateral with large cysts throughout the medulla and cortex and associated with ureteral abnormalities. Different from medullary cystic disease or polycystic kidneys.

multilobular liver - More than four liver lobes.

***multiple epiphysealis dysplasia** - See dysplasia epiphysealis multiplex.

myelocele - Protrusion of spinal cord substance through a defect in the bony spinal canal.

myelomeningocele - Spina bifida of the lumbar region. Exposure of the cord and meninges through a defect in the vertebral canal (spinal cord) [syn.: meningomyelocele].

myeloschisis - Congenital cleft of the spinal cord, owing to failure of the neural plate to form a complete tube.

N

nail-patella syndrome - Involvement of the head of the radius, hypoplasia or absence of the patellae, posterior iliac spurs, and dystrophy of the nails. [syn: arthro-onychodysplasia]

***neural tube defect** - A general term for a number of defects which are presumed to have a common origin in the failure of the neural tube to develop properly during embryogenesis. Conditions include, among others: anencephalus, craniorachischisis, encephalocele, iniencephaly, mengocele, meningoencephalocele, meningohydroencephalocele, meningomyelocele, myelocele, myelomeningocele, rachischisis, spina bifida (aperta, occulta and cystica), dermal sinus, and neuroenteric cyst. Some neural tube defects are associated with hydrocephalus.

***nevus** - A general term for a number of circumscribed stable malformations of the skin, and occasionally of the oral mucosa, which are not due to external causes. The excess (or deficiency) of tissue may involve epidermal, connective tissue, adnexal, nervous, or vascular elements. The malformation may be pigmented (yellow, pink, red, blue, brown or black), and have hair growing from it, or around the edges.

nodulated rib - A rib with an enlarged segment along its axis.

nonossified vertebral body - Failure of a ertebral body to ossify.

***Nuck's diverticula** - A diverticulum of the peritoneal membrane extending into the inguinal canal, accompanying the round ligament in the female, or the testis in its descent into the scrotum in the male (processus vaginalis testis); usually completely obliterated in the female. [syn: canal of Nuck, Nuck's canal, Nuck's diverticulum, or Nuck's hydrocele and Nuck's processus vaginalis peritonei]

nulliparous - Having never given given birth to a viable infant.

O

OAV syndrome - See Goldenhar's syndrome.

***oculoauricular dysplasia** - See Goldenhar's syndrome.

***oculoauricularvertebral (OAV) dysplasia** - See Goldenhar's syndrome.

oestrus - See estrus.

oestrous cycles - See estrous cycles.

oligoamnios - See oligohydramnios

oligodactyly - Fewer digits than normal (syn.: ectrodactyly, perodactyly). Also see dactyly.

oligohydramnios - Deficiency in the amount of amniotic fluid (syn.: oligoamnios)

oligosyndactyly - Fewer digits than normal with fusion of one or more. Also see dactyly.

***omphalocel or omphalocele** - Protrusion, at birth, of part of the intestine through a large defect in the umbilical ring (umbilicus), the protruding bowel being covered only by a thin transparent membrane composed of amnion and peritoneum. [syn.: umbilical hernia, exomphalos].

ophthalmia - Related to the eyes.

organogenesis - Period of organ formation.

ossification centers - Centers where ossification of bone is initiated. All bones in rodents should be ossified in a 20-day old fetus; the following sites are grouped into categories and usually are observed in a normal 20-day fetus.

Skull

Auditory ossicle	Nasal
Basisphenoid	Occipital/ exoccipital
Frontal	Parietals
Hyoid	Premaxillae
Interparietal	Presphenoid
Lacrimal	Squamosal
Mandibles	Tympanic Bullae
Maxillae	

Vertebrae	No. of Centra
Cervical	7
Thoracic	13
Lumbar	6
Sacral	4

Ribs	Sternum
13 pairs	6 ossification centers

Forepaw	Hind paw
Metacarpals #2, 3, 4, and 5	Metatarsals #2, 3, 4, and 5
Proximal phalanges #3 and 4	Distal phalanges #1, 2, 3, 4, and 5
Distal phalanges #1, 2, 3, 4, and 5	

***osteitis fibrosa disseminata** - See Albright-McCune-Sternberg syndrome.

***osteochondrodystrophia deformans** - See Morquio's syndrome..

***osteodystrophy** - Defective bone formation.

***osteogenesis imperfecta** - An inherited condition, usually transmitted as an autosomal dominant trait, in which the bones are abnormally brittle and subject fractures. [syn: fragilitas ossium, brittle bones, osteopsathyrosis, and hypoplasia of the mesenchyme]

***osteopetrosis** - Osteosclerotic bone disorder characterized by abnormally dense bones, probably due to defective resorption. In the autosomal recessive form occurring in infancy or childhood; the proliferation of the bone obliterates the bone cavity causing anemia, and the nerve foramina of the skull, causing compression of the cranial nerves, which may result in blindness and deafness. The benign autosomal dominant form occurs in adolescence or adulthood. Fractures are common in both forms. (associated with brittle bones that are prone to fracture, dwarfism, anemia, deafness, and hepatosplenomegaly.) [syn: Albers-Schoenberg disease, ivory bones and marble bones]

***ostepoikilosis** - A hereditary condition characterized by the presence of multiple sclerotic foci in the ends of long bones scattered stippling in round and flat bones, usually without symptoms and diagnosed fortuitously by x-ray examination. It is transmitted as an autosomal dominant trait.

***osteopsathyrosis** - See osteogenesis imperfecta.

***ostium primum** - An opening in the lowest aspect of the septum of the embryonic head.

***ostium secundum** - An opening high in the septum primum of the embryonic heart, approximately where the foramen ovale will be later.

***ostium secundum defect** - See atrial septal defect.

otocephaly - Fetus lacking lower jaw and having ears united below the face.

otocleisis - Closure of the auditory canals.

otorrhagia - Hemorrhage from the ear.

otorrhea - Discharge from the ear, especially a purulent one.

ovaries, hyperemic - Ovaries engorged with blood, generally due to hormonal stimulation. The term is also used to describe an ovarian condition in a bioassay for LH.

***overriding aorta** - A congenital anomaly occurring in tetralogy of Fallot, in which the aorta is displaced to the right so that it appears to arise from both ventricles and straddles the ventricular septal defect.

ovulation time - See estrous cycles.

beagle dog:	1-3 days after onset of estrus.
hamster:	8 to 12 hours after onset of estrus.
mouse:	2 to 3 hours after onset of estrus.
rabbit:	10 to 11 hours post-coitus(m) (induced).
rat:	8 to 10 hours after onset of estrus.
Rhesus monkey	11-14 days after onset of menses.

oxycephaly - Abnormally high, peaked, or conically shaped skull (termed as "domed head" by some investigators). [syn: acrocephaly, hypsicephaly, turricephaly, and steeple head or skull]

P

pachyotia - Abnormal thickness of the auricles of the ears.

palatoschisis - Cleft palate.

palpebral - An eyelid. Pertaining to eyelids.

parasitic twins - One incompletely developed fetus attached to a completely developed fetus.

***paratrophic** - Requiring living material or complex matter for food.

paresis - Slight or incomplete paralysis of limbs.

parity - State relative to having borne one or more offspring.

parous - Having borne one or more viable offspring.

Patau's syndrome - Holoprosencephaly (failure of cleavage of the prosencephalon into hemispheres or lobes) due to an aberration of the autosomes of the D group, in which central nervous system defects are associated with mental retardation, along with cleft lip and palate, polydactyly, dermal pattern anomalies and abnormalities of the heart, viscera, and genitalia. [syn: trisomy D syndrome and trisomy 13 syndrome]

patent ductus arteriosus - Failure of the ductus arteriosus to close soon after birth (connection between the aorta and pulmonary artery, resulting in abnormal recirculation of arterial blood through the lungs).

***patent ductus arteriosus reversed** - Patent ductus arteriosus with obstruction of the small vessels of the lungs, the direction of flow being from the pulmonary artery to the aorta, resulting in the return of venous blood to the systemic circulation with consequent cyanosis.

patent interventricular septum - Opening in the wall dividing the right and left ventricle of the heart.

patent ovale foramen - See foramen ovale. [Syn.: foramen ovale cordis].

pectoral girdle - Bones of the chest and thorax.

*pectus carinatum - Undue prominence of the sternum. [syn: chicken or pigeon chest]

*pectus excavatum - Undue depression of the sternum. [syn: funnel breast or chest]

pelvic girdle - The caudal portion of the trunk of the body, forming a basin bounded ventrally and laterally by the hip bones and dorsally by the sacrum and coccygeal vertebrae.

pelvic kidney - The kidney is displaced downward into the pelvic region.

pelvis - Any basin-like structure (e.g. the renal pelvis). Also applied to the bony pelvis formed by the sacrum, coccyx, ilium, pubis and ischium, bones that form the pubic and sciatic arches and hip.

*penile hypospadias - Hypospadias in which the urethral opening lies between the glandular sulcus and the junction of the penis and scrotum. [syn: secondary hypospadias]

*penoscrotal hypospadias - Hypospadias in which the urethral orifice is at the junction of the penis and scrotum; it may be associated with congenital chordee.

penineal hypospadias - Hypospadias in which the rudimentary penis is often engulfed by a bifid scrotum. The extreme form is called pseudovaginal hypospadias.

*penoscrotal hypospadias - Hypospadias in which the urethral orifice is at the junction of the penis and scrotum; it may be associated with congenital chordee.

*pentalogy of Fallot - The four defects included in the tetralogy of Fallot, occurring in association with patent foramen ovale or atrial septal defect.

*perineal hypospadias - Hypospadias with anomalous development of the genitalia, the rudimentary penis often being engulfed by overlying bifida scrotum. With perineal hypospadias the urethra opens in an area between the testicles proximal to the anus. [syn: pseudovaginal hypospadias in the extreme form]

pero- - Maimed; means deformed when used in combination with another term.

perobrachius - Deformed arms and forearms.

perodactyly - Fewer digits than normal [syn.: oligodactyly, ectrodactyl].

peromelia - Severely defective extremities including absence of hand/foot.

persistent common truncus - See persistent truncus arteriosus.

persistent ductus arteriosus - Persistent patency of the a fetal blood vessel connecting a pulmonary artery directly to the descending aorta. Normally patent prior to birth in the rat.

persistent truncus arteriosis - Aortic and pulmonary arteries fail to separate during development resulting in a common outflow tract for both vessels; always associated with ventricular septal defect.

*pes cavaus - Exaggerated height of the longitudinal arch of the foot, present from birth or appearing later because of contractures or disturbed balance of the muscles. When associated with clawing of the toes, it may be referred to as clawfoot.

*pes planovalgus - See pes planus.

*pes planus - The commonest form of flatfoot, in which one or more arches of the foot have flattened out due to insufficient tendon support. It is often hereditary. [syn: hypermobile flatfoot, pes planovalgus, and pes valgus]

*pes valgus - One form of flatfoot, in which the foot is rigid, and the position of the bones relative to each other has been altered with lowering of the longitudinal arch to produce a "rocker bottom" deformity. This deformity is caused by a primary dislocation of the talonavicular joint locking the foot into a plantar flexed position. [syn: hypermobile flatfoot, verticaltalus or rocker bottom flatfoot] Also see pes planus.

Peutz-Jeghers syndrome - Gastrointestinal polyposis (usually hamartomas of the small bowel) associated with excessive melanin pigmentation of the skin and mucus membranes.

*phakomatosis - An ophthalmologic term for any of four hereditary syndromes: (neurofibromatosis, tuberous sclerosis, encephalotrigeminal angiomatosis, and cerebroretinal angiomatosis) characterized by disseminated hamartomas of the eye, skin and brain.

phalanges - Pertaining to fingers and toes.

*pharyngeal pouch - A lateral diverticulum of the pharynx that meets a corresponding groove in the ectoderm, forming a closing plate.

philtrum - The groove at the median line of the upper lip.

*phocomelia - A developmental anomaly characterized by absence of the proximal portion of a limb or limbs, the hands or feet being attached to the trunk of the body by a single small irregularly shaped bone.

pica - Depraved appetite. Excessive eating of feces.

Pierre-Robin syndrome - Micrognathia in association with cleft palate, and glossoptosis, and absent gag reflex.

*pigeon chest - See pectus carinatum.

*pilonidal sinus - A supporting sinus containing a tuft of hair, occurring chiefly in the coccygeal region, but also in other regions of the body.

pinna - Auricle. Ear projecting outside of the head.

pinna unfolding -

mouse - 4.5 to 5 days.
rat - 5.0 to 6.5 days.

placenta - The organ of metabolic interchange between mother and fetus. The embryonic portion is derived from an outermost embryonic membrane (chorion frondosum), and a maternal portion formed by a modification of the uterine mucosa (decidua basalis) in which the fetal chorionic villi containing fetal capillaries are implanted. They are classified in several ways; based on the maternal and fetal tissue in contact with each other; based on the proportion of the fetal surface area that is in fact placentacious; and based on the amount of fetal loss at birth. Thus, bovine placenta is epitheliochorial, cotyledonary and nondeciduate. Also see the following individual types of placentae.

*plagiocephaly - An unsymmetrical and twisted condition of the head, resulting from irregular closure of the cranial sutures.

platy- - Broad, flat.

platyglossia - Broad, flat tongue.

platypodia - Lateral flattening of the foot. Also see Talipes planus, which means flat foot where entire sole touches the ground.

Poland's anomaly or syndrome - Unilateral absence of the sternocostal head of the pectoralis major muscle and ipsilateral syndactyly.

***polycystic kidney** - A hereditary congenital condition characterized by bilateral multiple renal cysts.

***polydactyly** - Having extra digits (fingers or toes). Also see dactyly, accessory fingers; accessory thumbs; skin tag.

***polydactyly (type A)** - See accessory fingers.

***polydactyly (type B)** - See skin tag.

polyestrous - Having more than one estrus cycle per mating season.

***polymicrogyria** - See microgyria.

***polyostotic fibrous dysplasia** - Manifestations including fibrous dysplasia of multiple bones, large pigmented cutaneous nevi with irregular margins and precocious sexual development from endocrine disturbance. [syn: disease: Albright's disease or syndrome; Albright-McCune-Sternberg]

***polyploidy** - The state of having more than two full sets of homologous chromosomes.

***porencephalia** - The presence of cysts or cavities in the brain cortex communicating by a "pore" with the arachnoid space.

postimplantation loss - Embryo/Fetal wastage.

Potter's syndrome - A rare condition combining a characteristic facial appearance (compression facies) with renal agenesis, or hypoplasia, and other defects.

premature delivery - Natural delivery of the products of conception prior to completion of the term of pregnancy. Generally refers to viable fetuses at birth.

presternum - See manubrium.

***primary hypospadias** - See balanic hypospadias.

proboscis - Cylindrical protuberance of the face, which, in cyclopia, represents the nose.

***processus vaginalis peritoni** - See Nuck's diverticulum.

prosencephalon - The anterior primitive cerebral vesical, or forebrain, dividing secondarily into telencephalon and diencephalon.

prosoposchisis - Fissure of the face.

prosopoanoschisis - Oblique fissure of the face.

prostration - Lying flat on the floor, powerless, extreme exhaustion, may or may not be conscious.

prune belly syndrome - A syndrome in which the lower part of the rectus abdominis muscle and the lower and the medial parts of the oblique muscles are absent, the bladder and ureters are usually greatly dilated, they are small and dysplastic, with hydronephrosis, and the testes are undescended.

pseudoencephaly - Congenital anomaly in which the vascular tissue replaces the bulk of the exposed brain.

pseudohermaphrodism - Sex glands of one sex but external genitalia of the opposite sex.

***pseudohermaphrodism, female** - Form in which affected individual is a genetic and gonadal female with partial masculinization.

***pseudohermaphrodism, male** - Form in which affected individual is a genetic and gonadal male with incomplete masculinization.

***pseudovaginal hypospadias** - See perineal hypospadias.

ptosis - Dropping upper eyelid(s) from paralysis of the third nerve or sympathetic innervation.

pulmonary valvular atresia - Absence of tissue or pathological closure of the pulmonary valve.

pyelectasia - Renal pelvic dilatation.

***pygopagus** - Conjoined twins consisting of two nearly complete individuals joined at the sacrum, so the two components are back to back.

pyometria - Pus within the uterus.

R

***rachischisis** - A type of spina bifida aperta with congenital fissures of the spinal column. It may be complete or partial. Also see craniorachischisis.

***ranual** - A cystic tumor beneath the tongue due to obstruction and dilatation of the sublingual or submaxillary duct or of a mucous gland.

Relative Teratogenic Index - Ratio between the minimum lethal dose for adults (LD01) and the minimum teratogenic dose (TD05) [Fabro et al., 1982]

renal papillae - The kidney consists of a cortex and medulla. The medullary substance forms pyramids, whose bases are in the cortex and whose apices, which are called papillae, project into the calices of the kidney.

renal pelvis - Funnel shaped expansion of the uppr end from the ureter into which the calices of the kidney open.

resorption - Loss of an embryo or fetus after implantation. Generally indicated by an implantation scar or metrial gland. Remnants of fetal tissue may be found at caesarean section in cases of fetal loss.

retina, folded - Congenital imperfection in the retina. The retina appears folded and usually has a fissure associated with the fold (sometimes an artifact due to fixation)

***retinocerebral angiomatosis** - See von Hippel-Lindau disease.

retrognathia - Repositioning of the jaws back of the frontal plane of the forehead.

reversed aorta - The aorta originates normally from the left ventricle, but is directed to the opposite side of the body (abnormal).

rhinencephalon - See arhinencephalia.

ringed aorta - The aorta forms a circular structure around the trachea.

rhinocephaly - A developmental anomaly characterized by proboscis-like nose above and between the eyes partially or completely fused.

rhypophagia - See coprophagia.

ribs - Thirteen pairs of thoracic ribs in rodents.

right-side descending aorta - Aorta descending on the right side instead of the normally on the left side.

***rocker bottom flat feet** - See pes valgus.

***Roger's disease** - A ventricular septal defect; the term is usually restricted to small, asymmetric defects.

rudimentary ribs - 14th ribs, less than half the length of 13th thoracic ribs (rodents).

runt - Usually refers to fetus weighing 25 percent less than the litter mean. Other definitions are used by laboratories, and include fetuses with decrease in weight of 2 standard deviations from the mean, and fetuses with graft versus host disease.

S

sacral vertebral arches and centra - There are 6 vertebral arches and centra.

supernumerary rib - See fourteenth rib.

***sacrococcygeal** - Pertaining to or located in the region of the sacrum and coccyx.

Salewski (method) - Macroscopic identification of implantation sites (metrial glands) in rodent uteri using 10% aqueous (v/v) ammonium sulfide solution.

scatophagy - See coprophagia.

schisis - Fissure.

sclerosis - Hardening of a tissue.

***scoliosis** - Lateral curvature of the normally straight vertical line of the spine. Also see kyphosis and lordosis.

scotoma - Blind spot (depressed vision) in the field of vision (may be normal or abnormal).

scrambled sternebrae - Sternebrae, ossified or not, appear on two sides of the central axis of the sternum.

***scrotal hypospadias** - Abnormal opening of the urethra onto the scrotum. [syn: tertiary hypospadias]

***secondary hypospadias** - See penile hypospadias.

Segment 1 studies - Food and Drug Administration's (Phase I) studies are one-generation studies of test compound effects on gondal function, estrous cycles, mating behavior, conception rates and early stages of development. It covers 60 days of treatment for males, 14 days of treatment for females prior to mating, followed by treatment through gestation, parturition, and weaning.

Segment II studies - Food and Drug Administration's (Phase II) studies involve test compound treatment of two species (rodent and non-rodent) from day 6 through major organogenesis; caesarean section is performed just prior to parturition and the conceptus is examined for gross, viseral, and skeletal abnormalities.

Segment III studies - Food Drug Administration's (Phase III) studies involve test compound treatment of the rat from gestational days 15 through the end of weaning (postnatal day 21). It determines effects on late fetal development, labor, delivery, lactation and neonatal survival and growth.

semen volume - In man, 30 percent comes from prostate and 60 percent from the seminal vesicles.

***septum primum** - A septum in the embryonic heart which divides the primitive atrium into right and left chambers.

***septum secundum** - A septum in the embryonic heart to the right of the septum primum; after birth it fuses with the septum primum to close the foramen ovale.

sex ratio - Ratio or percentage of each sex against the total number of offspring calculated at birth and at 4, 7, 14, and 21 days of age.

sexual maturity - (Depends on strain, nutrition, and other factors). Also see breeding age which is not the same.

hamster - 42 to 54 days
mouse - 28 days
rat - 46 to 53 days
rabbit - 120 to 240 days
guinea pig - 84 days

sigmoid kidney - Fusion of lower pole with the upper pole of the contralateral kidney.

sirenomelia - Fusion of the legs and feet. In Greek mythology, a siren or sea-nymph, half woman and half fish. In later times a mermaid.

situs inversus - Lateral transposition of the thoracic and abdominal viscera; usually affecting the heart.

Sjogren-Larssen syndrome - Congenital oligophrenia, ichthyosis and spastic pyramidal symptoms.

***skin tag** - A rudimentary appendage or pendulous growth of tissue without a bone. Postaxial skin tags may be called polydactyly (type B).

skull, bone islands - Accessory skull bones; considered as variations within the continuum of developmental stages of fetal craniofacial osteogenesis.

***smell-brain** - See arhinencephalia.

spermatogenesis - Formation of sperm.

spermatogenic cycle -

1. spermatocytogenesis - Spermatogenic growth phase, consists of mitotic (proliferative) and meiotic (reductive) divisions.

mouse - 21.3 days
rat - 27.8 days
man - 39.2 days

2. spermiogenesis - Complete development of spermatids into sperm.

mouse - 12.5 days
rat - 20.6 days
man - 21.0 days

3. post-testicular ductular transit time

mouse - 8 days
rat - 14 days
man - 21 days

spermiogenesis - The second stage in the formation of spermatozoa in which the spermitids transform into spermatozoa.

Development stages: spermatogonia → primary spermatocytes
→ secondary spermatocytes → spermatids → spermatoza.

***spider finger** - See arachnodactyly.

spina bifida - A developmental anomaly characterized by defective closure of the bony encasement (vertebral arches) of the spinal cord, through which the cord and meninges may or may not protrude. Also see spina bifida aperta, spina bifida cystica, and spina bifida occulta.

***spina bifida aperta** - Spina bifida in which the spinal canal is open with neural tissue exposed and no skin cover. Also see rachischisis.

spina bifida cystica - Spina bifida with skin cover and protrusion through the defect of a cystic swelling involving the meninges (meningocele), spinal cord (myelocele), or both (meningomyelocele). About 75% of the lesions are both.

***spina bifida occulta** - Spina bifida with no protrusion or cyst, with the lesion covered by skin. The skin usually has hair over the lesion, and may be pigmented or dimpled.

***spondyloepiphyseal dysplasia** - See Morquio's syndrome.

***spondyloisthesis** - Forward displacement of on one vertebra over another, usually of the fifth lumbar over the body of the sacrum, or of the fourth lumbar over the fifth.

spondyloschisis - Congenital fissure of vertebral arches. [syn: rachischisis]

***sprengel's deformity** - Congenital elevation of the scapula, due to failure of descent in fetal life.

staphyloschisis - Cleft palate. Bifid uvula with or without the soft palate.

steeple head or skull - See oxycephaly.

stenosis - Constriction, narrowing.

sternum - Breast bone; Consists of sternebrae in newborn rodents (5th sternebrae is last one to ossify).

stillborn - Death in utero; born dead.

***stippled epiphyses** - See Conradi's disease.

structural alteration - Structural alterations or delays in development that include both malformations and variations.

supernumerary - Sum of extra and rudimentary structures.

supernumerary rib - Extra rib. See also fourteenth rib.

supernumerary kidney - Third kidney (usually small).

symphalangia - Fusion of two adjacent phalanges. Web-fingers or web-toes.

symphalangism - Syndactylism (webbing together of fingers or toes, dactylion, dactylosymphysis) or ankylosis (stiffening or fixation) of a finger or toe joint.

synchilia - Adhesion of lips. Atresia of the lips.

syndactyly - Webbed or fused digits (see dactyly).

syndesmochorial placenta - A type of placenta characterized by an endometrial attachment to the chorion with limited destruction of the endometrial epithelium. Formally thought to be characteristic of sheep, cattle, and goats , which are now known to epitheliochorial placentae.

synophthalmia - Partial or complete fusion of the eyes . [syn: cyclopia and synopsia]

synopsia - Fusion of the eyes.

synopsy - The abnormal suggestion of types of the human face or figure by the various numerals.

synphalangy - Fusion of two or more phalanges.

synotia - Fusion or abnormal approximation of the lobes of the ears, usually associated with absence or incomplete development of the lower jaw.

T

talipes (club foot) - Congenital deformity of the foot; a foot that has grown in a twisted manner resulting in an abnormal shape or position. Talipes are divided into: t. equinus, t. varus, t. valgus, t. calcaneus.

***talipes calcaneovalgus** - A deformity of the foot in which the heel is turned outward from the midline of the body and the anterior part of the foot is elevated.

***talipes calcanevorus** - A deformity of the foot in which the heel is turned toward the midline of the body and the anterior part is elevated.

***talipes equinovarus** - A deformity of the foot in which the heel is turned inward from the midline of the leg and the foot plantar flexed.

talus - Second largest bone of the ankle.

*teras - A grossly malformed fetus or infant (Greek: teras - a monster).

*terata - Plural of teras. Permanent structural or functional anomalies that are considered to be adverse to the organism.

*teratism - An anomaly of formation or development.

*teratogen - An agent or factor that causes physical defects in the developing (embryo) organism.

teratogenicity - The power, property or the ability to produced terata. Used most frequently by government agencies to facilitate reference to teratogenicity, carcinogenicity and mutagenicity. The term is not included in dictionaries, but never the less joins the ranks of other idiomatic expressions such as: offloading, on track, production volume, and terminate with extreme prejudice.

teratology - The study of abnormal development. The division of embryology and pathology, considering congenital malformations.

*tertiary hypospadias - See scrotal hypospadias.

*tetralogy of Fallot - Cardiac defects consisting of pulmonary stenosis, interventricular septal defect, dextroposition of the aorta so that it overrides the interventricular septum and receives venous as well as arterial blood, and right ventricular hypertrophy.

*thanatophoric - Dwarfism resulting in death due to respiratory insufficiency in the newborn period. Clinically, these are exaggerated achondroplastic dwarfs with flat vertebral bodies and short, thick, curved tubular bones.

thoracic arches and centra - There are 13 cervical arches and centra. More or less than 13 thoracic arches are considered to be a variation, but less has been considered to be a malformation.

*thoracopagus - Conjoined twins consisting of two nearly complete individuals joined in or near the sternal region, so the two components are face to face.

thoracoschisis - Congenital fissure of the thorax and possibly lung herniation.

*thyrolingual duct - See ductus thyroglossus. [syn: thyroglossal duct]

*tongue-tie - Abnormal shortness of the frenum of the tongue, resulting in limitation of the motion. [syn: ankyloglossia and linguia frenata]

*total anomalous pulmonary venous return - A congenital heart defect where all the pulmonary veins drain into the right atrium.

tracheal stenosis - Constriction of tracheal lumen.

tracheoesophageal fistula - Esophagus connecting with trachea.

*tracheomalacia - Softening of the tracheal cartilages.

transposition, vessels - e.g., Aorta originates from right instead of left ventricle.

*transposition of great vessels - A congenital cardiovascular malformation in which the aorta arises entirely from the right ventricle and the pulmonary artery from the left ventricle so that the venous return from the peripheral circulation is recirculated by the right ventricle, via the aorta, to the systemic circulation without being oxygenated in the lungs.

Treacher-Collins syndrome - A hereditary disorder occurring in two forms: the complete form (Franceschetti's syndrome) is characterized by antimongoloid slant of the

palpebral fissures, coloboma of the lower lid, micrognathia and hypoplasia of the zygomatic arches, and microtia. It is transmitted as an autosomal dominant trait. The incomplete form (Treacher-Collins syndrome) is characterized by the same anomalies in less pronounced degree. It occurs sporadically, but an autosomal dominant mode of transmission is suspected. [syn: mandibulofacial dysostosis]

triatrial heart - See Cor triatriatum.

***trigonocephaly** - A deformity of the head characterized by sharp angulation ventrad of the squamous portion of the frontal bones at the site of the suture between them.

trilocular - Having three compartments or cells.

***triploidy** - The presence in humans of 69 chromosomes, or three full sets, a frequent finding in abortuses.

trisomy D syndrome - See Patau's syndrome.

trisomy E syndrome - See Edwards' syndrome.

trisomy 13-15 syndrome - See Patau's syndrome.

trisomy 16-18 syndrome - See Edwards' syndrome.

trisomy 21 syndrome - See Down's syndrome.

***truncus arteriosus, persistent** - A single trunk arising from the heart, receiving blood from both ventricles and supplying blood to the coronary, pulmonary, and systemic circulations.

truncus communis - Common aortic and pulmonary channel.

Turner's syndrome - See gonadal dysgenesis.

turricephaly - See oxycephaly.

tympanic bullae - Symmetrical pair of ossification centers located on either side of the skull, inside the brain case just behind the jaw bone.

twins - See conjoined twins, monozygotic twins and parasitic twin.

U

umbilical hernia - Protrusion of the bowel or omentum at the navel. [syn. omphalocele and exomphalos]

unilateral sternebral ossification - Only one half the sterenbra is ossified. [Syn: hemisternebra]

unilobular lung - Condition in which the lung consists of one lobe instead of several (the right lung of the rat has four lobes).

***urachus** - A canal in the fetus that connects the bladder with the allantois.

ureters - Tube that connects kidneys with the urinary bladder.

***urticaria pigmentosa** - Mastocytosis manifested as persistent pink to brown macules or soft plagues of various sizes, the irritation of which results in localized pruritus and urtication (Darier's sign).

V

vaginal opening -
mouse - 30 to 40 days
rat - 35 to 50 days

variation - A divergence beyond the usual range of structural constitution, which may not adversely affect survival, development, or function.

***ventricular septal defect** - A cardiac anomaly in which there is persistent patency of the ventricular septum in either the muscular or fibrous portions.

vertebrae ossification sites - See Ossification Centers.

***vertical talus** - See pes valgus.

viability status - For rodent reproduction studies: Differentiation between liveborn, but dead immediately after birth and stillborn (death in utero) by removing the lung and immersing in water (float = liveborn).

***Von Hippel-Lindau disease** - Phakomatosis (four hereditary syndromes; neurofibromatosis, tuberous sclerosis, encephal-trigeminal angiomatosis, and cerebroretinal angiomatosis) characterized by congenital angiomatosis of the retina and cerebellum. [syn: cerebroretinal or retinocerebral angiomatosis and Lindau-von Hippel disease]

W

wavy rib - Ribs bent in more than one direction and/or the angle of the bend may change, i.e., wavy. May be attributed to differential development of ribs and intercostal muscles.

weight gain, corrected - Maternal weight gain during gestation minus gravid uterine weight.

weight, Maximum Stunted (MSW) - MSW < or = (mean group fetal weight with the smallest fetal weight omitted) X 0.666. If the omitted fetus weighs less than or equal to the MSW, it is designated as a "calculated stunted."

weight, post weaning - Post weaning body weight in grams for selected strains and species:

Age (in days)	Males	Females
	Sprague-Dawley Rat	
18-20	35-50	35-50
21-30	50-90	50-85
31-40	90-170	85-160
41-50	170-250	160-200
51-60	250-320	200-225
61-70	320-370	225-250
>70	>370	>250
	Fisher 344 rat	
18-20	<35	<35
21-30	35-50	35-50
31-40	50-95	50-75
41-50	95-125	75-100
51-60	125-175	100-125
61-70	175-200	125-150
>70	>200	>150
	Mouse	
18-20	10-12	10-12
21-30	12-25	12-18
31-40	25-30	18-23
41-50	30-35	23-30
51-60	>35	>30

***Wolffian duct** - An embryonic duct initiated in association with rudiments of the pronephric kidney, taken over as ducts by the reproductive system in males and into vestigial structures in females. [syn: ductus Wolffi, duct of Wolff, ductus mesonephricus, Leydig's (mesonephric) duct and canal of Oken]

Wolff-Hirschorn syndrome - Deletion of chromosome 14 in humans.

X

xiphoid sternebrae - Sternebrae shaped like a sword; usually refers to the 6th sternebrae. [syn: Xiphisternum]

***xiphopagus** - Symmetrical conjoined twins fused at the sternum in the region of the xiphoid process.

BIBLIOGRAPHY

Adams, C. E. 1961. Artificial insemination. In: The semen of animals and artificial insemination. ed. J. P. Maude. England.

Arey, L. B. 1965. *Developmental Anatomy: A Textbook and Laboratory Manual of Embryology*. W. B. Saunders Co., Philadelphia.

Benirschke, K., F. M. Garner, and T. C. Jones, *eds*. 1978. *Pathology of Laboratory Animals*. Springer Verlag.

Blood, D. C., and V. P. Studdert. 1988. *Baillière's Comprehensive Veterinary Dictionary*. Baillere Tindall Press, Philadelphia.

Chang, M. C. 1958. Capacitation of rabbit spermatozoa in the uterus with special reference to the reproductive phases of the female. *J. Endocrinol*. 63:619-628.

Crump, K. S. 1984. A new method for determining allowable daily intakes. *Fundam. Appl. Toxicol*. 4:854-871.

Dawson, A. B. 1926. A note on the staining of the skeleton of cleared specimens with Alizarin Red S. *Stain Technology* 1:123.

Dorland's Illustrated Medical Dictionary, 25th ed. 1976. W. B. Saunders Co., Philadelphia.

Engler, R., and J. A. Quest. 1988. Review of the Office of Pesticide Programs Current Toxicology Guidelines for the Testing of Pesticides. *Toxicology and Industrial Health*. 4(3):323-329.

Farris, E. J., and J. J. Griffith, *eds*. 1967. *The Rat in Laboratory Investigation*. Hafner Publishing Co., New York.

Gad, S. C. and C. P. Chengelis *eds*. 1988. Routes, Formulations and Vehicles. Ch. 10, Appendix C In: *Acute Toxicology Testing Perspectives and Horizons*. The Telford Press, Caldwell, New Jersey.

Gad, S., and C. Weil. 1982. Statistics for Toxicologists. Ch. 9. In: *Principles and Methods of Toxicology*. A. W. Hayes, *ed*. Raven Press, New York.

Gad, S., and C. S. Weil. 1988. *Statistics and Experimental Design for Toxicologists*. The Telford Press, Caldwell, New Jersey.

Gibson, J. P., R. E. Staples, and J. W. Newberne. 1966. Use of rabbit in teratogenicity studies. *Toxicol. Appl. Pharmacol*. 9:398-408.

Green, E. L. 1962. Quantitative genetics of skeletal variations in the mouse. II. Crosses between four inbred strains (C3H, DBA, C57BL, BALB/c). *Genetics*. 47:1085-1096.

Hafez, E. S. E., *ed*. 1970. *Reproduction and Breeding Techniques for Laboratory Animals*. Lea and Febiger, Philadelphia.

Hafez, E. S. E. *ed*. 1980. *Reproduction in Farm Animals*. Lea and Febiger, Philadelphia.

Harbison, R. D. 1980. Teratogens. Ch. 8. In *Caserett and Doull's Toxicology: The Basic Science of Poisons*. 2nd Ed. Douell, J., C. D. Klassen, and M. O. Amdur, *eds*. Macmillan Publishing Co., Inc. New York.

Manson, J. M., W. Zemik, and R. D. Costlow. 1984. Teratology Test Methods for Laboratory Animals. in: *Principles and Methods of Toxicology*. Hayes, A. W., *ed*. Raven Press, New York.

Jelovsek, F. R., D. R. Mattison, and J. P. Young. 1990. Eliciting principles of hazard identification from experts. *Teratology*. 42:521-533.

Kavlock, R. J., N. Chernoff, and E. H. Rogers. 1985. The effect of acute maternal toxicity on fetal development in the mouse. *Teratogenesis, Carcino. and Mutagen*. 5:3-13.

Kimmel, C. A. and E. Z. Francis. 1990. Proceedings of the workshop on the acceptability and interpretation of dermal developmental toxicity studies. *Fundam. Appl. Toxicol*. 14:386-398.

Kimmel, C. A., and D. W. Gaylor. 1988. Issues in qualitative and quantitative risk analysis for developmental toxicology. *Risk Anal*. 8:15-20.

Kimmel, C. A., and C. J. Price. 1990. Developmental toxicity studies. In: Arnold, D.L., H. C. Grice, D. R. Krewski, *eds*. *Handbook of In Vivo Toxicity Testing*. Academic Press, San Diego, CA.

Kimmel, C. A., and J. G. Wilson. 1973. Skeletal deviations in rats: Malformations or variations? *Teratology*. 8:309-316.

Mattison, D. R. 1991. Physiological alterations during pregnancy: impact on toxicokinetics. *Fund. Appl. Toxicol.* 16(4):215-218.

Middle Atlantic Reproduction and Teratology Association (MATRA). Glossary of terms.

Murphy, S. D. 1980. Pesticides. Ch. 16. In *Caserett and Doull's Toxicology: The Basic Science of Poisons.* 2nd Ed. Douell, J., C. D. Klassen, and M. O. Amdur, eds. Macmillan Publishing Co., Inc. New York.

National Research Council. 1983. Risk assessment in the federal government: Managing the process. National Academy Press, Washington D. C., p. 159. ISBN 0-309-03979-7

National Research Council. 1989. Biologic markers in reproductive toxicology: Subcommittee on reproductive and neurodevelopmental toxicology committee on biologic markers. National Academy Press, Washington D. C. ISBN 0-309-03979-7

Nau, H. 1991. Pharmacokinetic considerations in the design and interpretation of developmental toxicity studies. *Fund. Appl. Toxicol.* 16(4): 219-221.

Palmer, A. K. 1977. Incidence of sporadic malformations, anomalies and variations in random bred laboratory animals. In: *Methods in Prenatal Toxicology.* Neubert, D., H. J. Merker, and T. E. Kwasigroch, *eds.* Georg Thieme Pub., Stutgart.

Sawin, P. B., and M. Gow. 1967. Morphogenetic studies of the rabit. XXXVI. Effect of gene and genome interaction on homeotic variation. *Anat. Rec.* 157:425-435.

Scott, W. J., R. L. Butcher, C. W. Kindt, and J. G. Wilson. 1972. Greater sensitivity of female than male rat embryos to acetazolamide teratogenicity. *Teratology.* 6:239-240.

Snedecker, G. W., and W. G. Cochran. 1989. *Statistical Methods,* 8th ed. Iowa State University Press, Ames, Iowa.

Sokal, R. R., and E. J. Rolf. 1981. *Biometry.* W. H. Freeman, San Francisco.

Staples, R. E. 1977. Detection of visceral alterations in mammalian fetuses. *Teratology.* 9:A37-A38.

Stedman's Medical Dictionary, 21st ed. 1966. The Williams and Wilkins Co., Baltimore.

Thomas, C., *ed.* 1981. *Tabor's Cyclopedic Medical Dictionary,* 14th ed. F. A. Davis Co., Philadelphia.

USEPA. October, 1982. *Pesticide Assessment Guidelines, Subdivision F, Hazard Evaluation: Human and Domestic Animals.* Office of Pesticides and Toxic Substances, Washington, DC. EPA-540/9-82-025. Available from NTIS, Springfield, VA.

USEPA. June, 1985. *Hazard Evaluation Division Standard Evaluation Procedure: Teratology Studies.* Office of Pesticide Programs, Washington, D. C. EPA-540/9-85-018. Available from NTIS, Springfield, VA.

USEPA, 1989. FIFRA Accelerated Reregistration Phase 3 Technical Guidance, Office of Pesticides and Toxic Substances, Washington, DC. EPA No. 540/09-90-078. Available from NTIS, Springfield, VA.

USEPA, 1991a. *Pesticide Assessment Guidelines, Subdivision F, Hazard Evaluation: Human and Domestic Animals,* Addendum 10, Neurotoxicity, Series 81, 82, and 83. March, 1991, EPA 540/09-91-123, PB 91-154617.

USEPA, 1991b. *Guidelines for Developmental Toxicity Risk Assessment.* December 5, 1991. Federal Register, Vol. 56, No. 234, pages 63798-63826.

Walton, A. 1927. Relation between density of sperm suspension and fertility as determined by artificial insemination of rabbits. *Proc. Roy. Soc. B.* 101:303-315.

Williams, R. H., *ed.* 1974. *Textbook of Endocrinology.* W. B. Saunders Company, Philadephia.

Wilson, J.G., 1965. Methods for administering agents and detecting malformations in experimental animals. In: Teratology: Principles and Techniques.,eds J.G. Wilson and J. Warkany. Univ. of Chicago Press.

Wilson, J.G. 1973. In: Environment and birth defects. ed., J.G. Wilson, Academic Press.

Woo, D.C. 1984. Control data of teratological studies in New Zealand White rabbits (A joint study by MARTA), in *Principles in Teratology,* Teratology Society.

Woo, D.C. and Hoar, R.M., 1982. Reproductive performance and spontaneous malformations in control New Zealand rabbits: A joint study by MARTA. Teratology 25, 82A.